PB-214 994

# THE FATE OF ABS IN SOILS AND PLANTS

Stephen A. Klein, et al

California University
Berkeley, California

October 1962

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# THE FATE OF ABS IN SOILS AND PLANTS

Stephen A. Klein
David I. Jenkins

P. H. McGauhey
Faculty Investigator

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Sanitary Engineering Research Laboratory
University of California
Berkeley, California

## Part I. REDUCTION BY SOILS

### INTRODUCTION

As an expanding population imposes increasing demands upon a relatively fixed water resource, waste water must inevitably appear as an increasingly important fraction of both surface and ground waters. The fate of such waste water components as ABS in the soil and their effects on plant life is therefore of vast importance in determining the feasibility of waste water reclamation by surface spreading or underground injection, the economics of waste water treatment, and the commercial future of the detergent itself.

Previous work (1) by the authors has shown that it is possible to reduce the ABS concentration of a sewage effluent to about 1 mg/l by induced frothing. Such a concentration is now widely recognized as the threshold of frothing of ABS and is the basis for regulatory "standards" or limits on the permissible concentration of ABS in the water environment. A concentration of 1 mg/l was therefore selected for studies of removal of ABS from percolating waters by biological degradation and adsorption on soils. To simulate both surface and underground conditions, experiments were run on both sterile and normally biologically active soils, and under saturated and unsaturated conditions. Concentrations up to 50 mg/l of ABS were utilized in experiments on the effect of the detergent on plant growth, reported in Part II of this paper.

## MATERIALS AND METHODS

### Feed Solution

The feed solution used in the experiments was a simulated sewage effluent containing ABS concentrations of the order of 1 mg/l. It consisted of BOD dilution water (2) containing sufficient Difco nutrient broth to give a BOD of 20 mg/l plus 50-mg/l chloride ion. ABS (about 1 mg/l) and $ABS^{35}$ (in tracer quantities) were added to 16 liters of feed solution in a 5-gallon carboy (Figure 1) which was then sterilized by autoclaving.

The ABS was prepared by the sulfonation of an alkane made up of alkylbenzenes representative of the types used by five major detergent manufacturers (California Research Corporation). For tracer work such ABS was labelled with $S^{35}$ by sulfonating the alkylbenzene with $H_2S^{35}O_4$.

### Soil Columns

Five soils as characterized in Table 1 were used in the investigation. All are typical agricultural soils in California collected from areas where sewage spreading has been projected or is practiced. Of the five, three are classified as sandy loams (Columbia, Hesperia, and Yolo), one is a fine sandy loam (Hanford), and one a sand (Oakley).

For percolation experiments the air-dried soils were packed into a 2.5-cm diameter glass column to a depth of 25 cm, overlying an underdrainage system, as shown in Figure 2. The soil column was then sterilized by autoclaving. In use it received the feed solution at a constant head, maintained by a Mariotte syphon.

### Analysis of Column Effluent

Samples of the column effluent were collected in graduated cylinders over timed intervals so that the rate of percolation could be

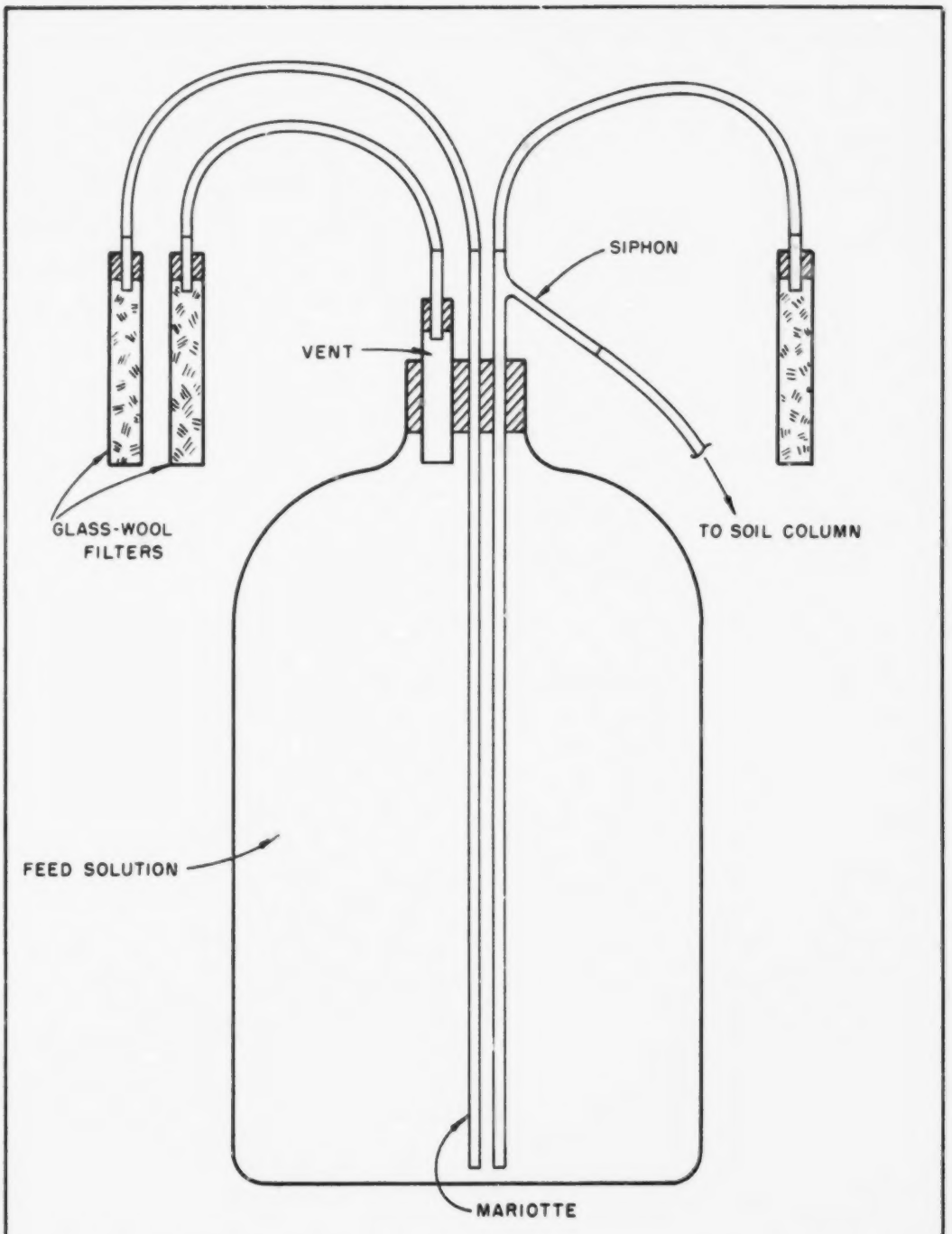


FIGURE 1. FEEDING ARRANGEMENT FOR SOIL COLUMN

TABLE 1

PHYSICAL AND CHEMICAL SOIL CHARACTERISTICS

| Soil | Parent Material | Soil Reaction | Sub-Surface Drainage | Particle Size, % | | | | | | Exchange Capacity, me/100g | Carbon, % | | Porosity, % | Density, g/ml |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Clay | Silt | Sand | | | | | | | | |
| | | | | | | Very Fine | Fine | Med. | Coarse | | Org. | $CO_3$ | | |
| Columbia | Mixed alluvium | Neutral | Good | 15.5 | 21.5 | 9.5 | 13.5 | 32 | 8 | 4.6 | 0.59 | 0.00 | 40 | 1.62 |
| Hanford | Granitic alluvium | Neutral | Good | 17.5 | 17.5 | 10 | 23 | 24 | 8 | 4.3 | 0.64 | 0.00 | 40 | 1.57 |
| Hesperia | Granitic alluvium | Neutral | Good | 19 | 15 | 1 | 20 | 34 | 11 | 7.0 | 0.28 | 0.00 | 39 | 1.57 |
| Yolo | Sedimentary alluvium | Neutral | Good | 23 | 19 | 13 | 27 | 17 | 1 | 11.6 | 0.06 | 0.00 | 49 | 1.34 |
| Oakley | Sandy, mixed alluvium | Slightly acid | Excessive | 12 | 1 | 9 | 35 | 41 | 2 | 2.3 | 0.05 | 0.00 | 41 | 1.63 |
| Well-Field Bailings | --- | --- | --- | 4 | 0 | 10 | 3 | 4 | 79 | 7.6 | 0.08 | 0.09 | 35 | 1.78 |

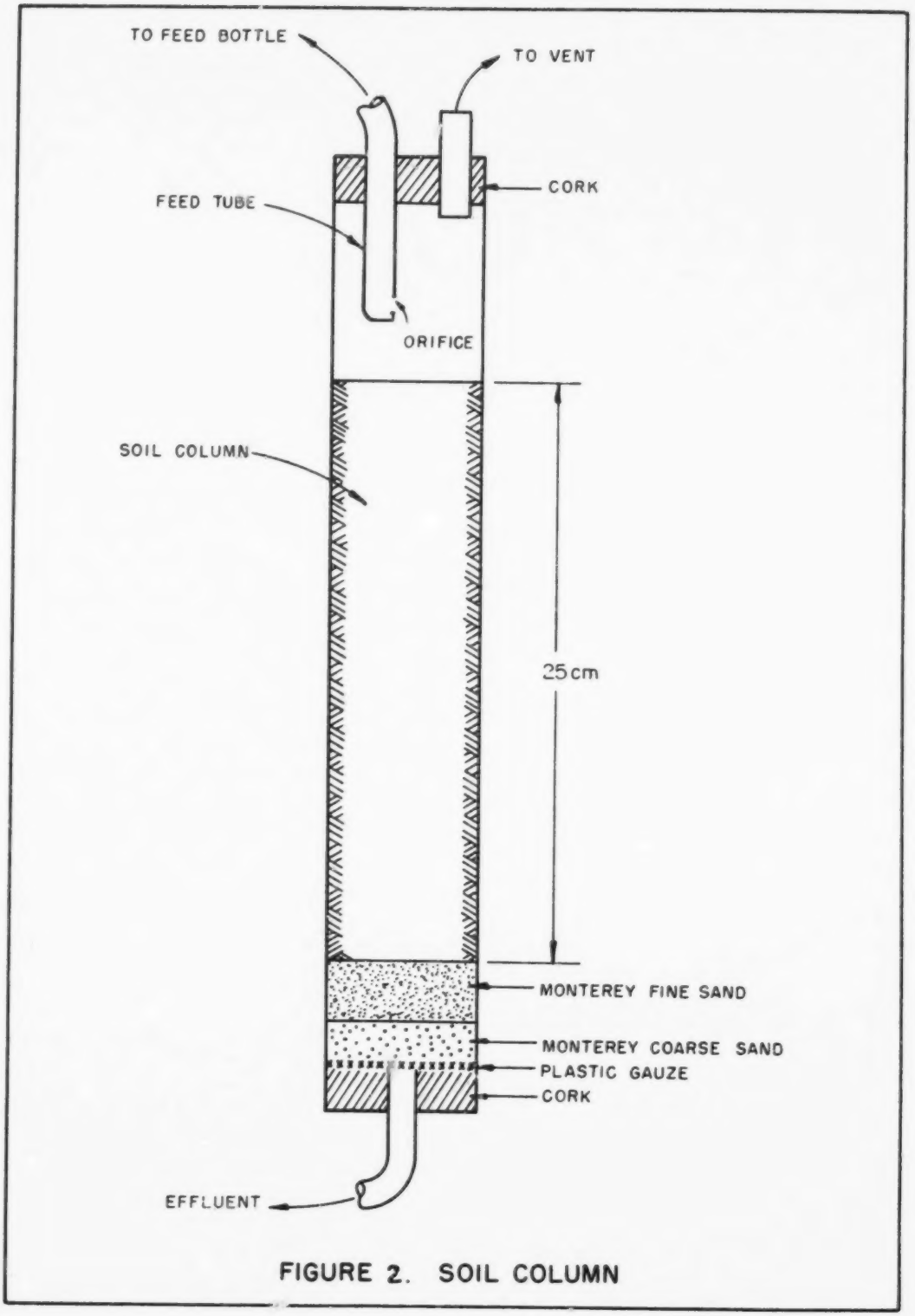


FIGURE 2. SOIL COLUMN

estimated. The effluent samples were analyzed for $Cl^-$ (2), total $S^{35}$, $SO_4$-$S^{35}$, and ABS-$S^{35}$ (3) when appropriate. The ABS carrier concentration in the feed was determined by the methyl green method (4). The breakthrough of ABS-$S^{35}$ and $S^{35}$ has been depicted in normalized graphical form using the $Cl^-$ curve to trace the hydraulic breakthrough.

Analysis of Soil

To determine the ABS adsorption it was necessary to analyze the soil itself for ABS-$S^{35}$ and $SO_4$-$S^{35}$. After draining the column, the soil was extruded and divided into six portions: Top-2.5 cm; 2.5 cm-5 cm; 5 cm-10 cm; 10 cm-15 cm; and 15 cm-25 cm. Samples of each section were extracted with hot ethanol (3) and analyzed for ABS-$S^{35}$ and $SO_4$-$S^{35}$, a correction being made for the amount of ABS-$S^{35}$ contained in the liquid remaining in the soil fractions.

EXPERIMENTAL RESULTS

Sterile, Saturated Soil Columns

The air in the soil column was completely displaced with $CO_2$ so that on introduction of feed solution saturated flow conditions were attained. During each of the column studies there were intervals when the columns were clamped off at the bottom, and allowed to rest, in a saturated condition. On resuming the flow through the column, the $S^{35}$ and ABS-$S^{35}$ content of the effluent is initially lower. The first liquid leaving the column is that which has been in contact with the soil column during the static contact period. Thus even though 100 per cent breakthrough may have been obtained under flow conditions, further ABS adsorption takes place during static contact. Although the degree of adsorption is so small that it cannot be observed when the column is being percolated

with ABS solution, the effect of these "static contact" periods is quite pronounced. A typical breakthrough curve showing the effect of static contact periods is shown in Figure 3.

In all soil columns the 100 per cent breakthrough of $S^{35}$ occurred much more slowly than the hydraulic breakthrough, although small amounts of $S^{35}$ began to appear in the effluent almost immediately (Table 2).

TABLE 2

BREAKTHROUGH OF $CL^-$ AND $S^{35}$ ON STERILE, SATURATED SOIL COLUMNS

| Soil | Pore Volumes for Breakthrough | | |
|---|---|---|---|
| | Chloride | $S^{35}$ Front | |
| | | 90 Per Cent | 100 Per Cent |
| Oakley | 1.7 | 12.3 | 23.4 |
| Yolo | 1.8 | 21.3 | 36.0 |
| Hesperia | 2.3 | 27.5 | 35.0 |
| Hanford | 2.4 | 34.5 | 60.0 |
| Columbia | 2.3 | 28.2 | 61.8 |

The greatest adsorption per liter of feed percolated occurred in the first liter throughput. Thereafter the amount adsorbed per liter rapidly decreased (Table 3). These values can be used to define the ABS-adsorbing capacity of each soil under sterile, saturated flow conditions in terms of loading in million gallons/acre foot. Thus, from Figure 4, it is estimated that after the infiltration of 6 million gallons/acre foot through Oakley sand, 98.5 per cent of the ABS applied would be discharged in the effluent. At this same loading for the other four soils, the percentage of influent ABS in the effluent would be: Hesperia, 88.5; Yolo, 87.5; Hanford, 81.0; and Columbia, 79.

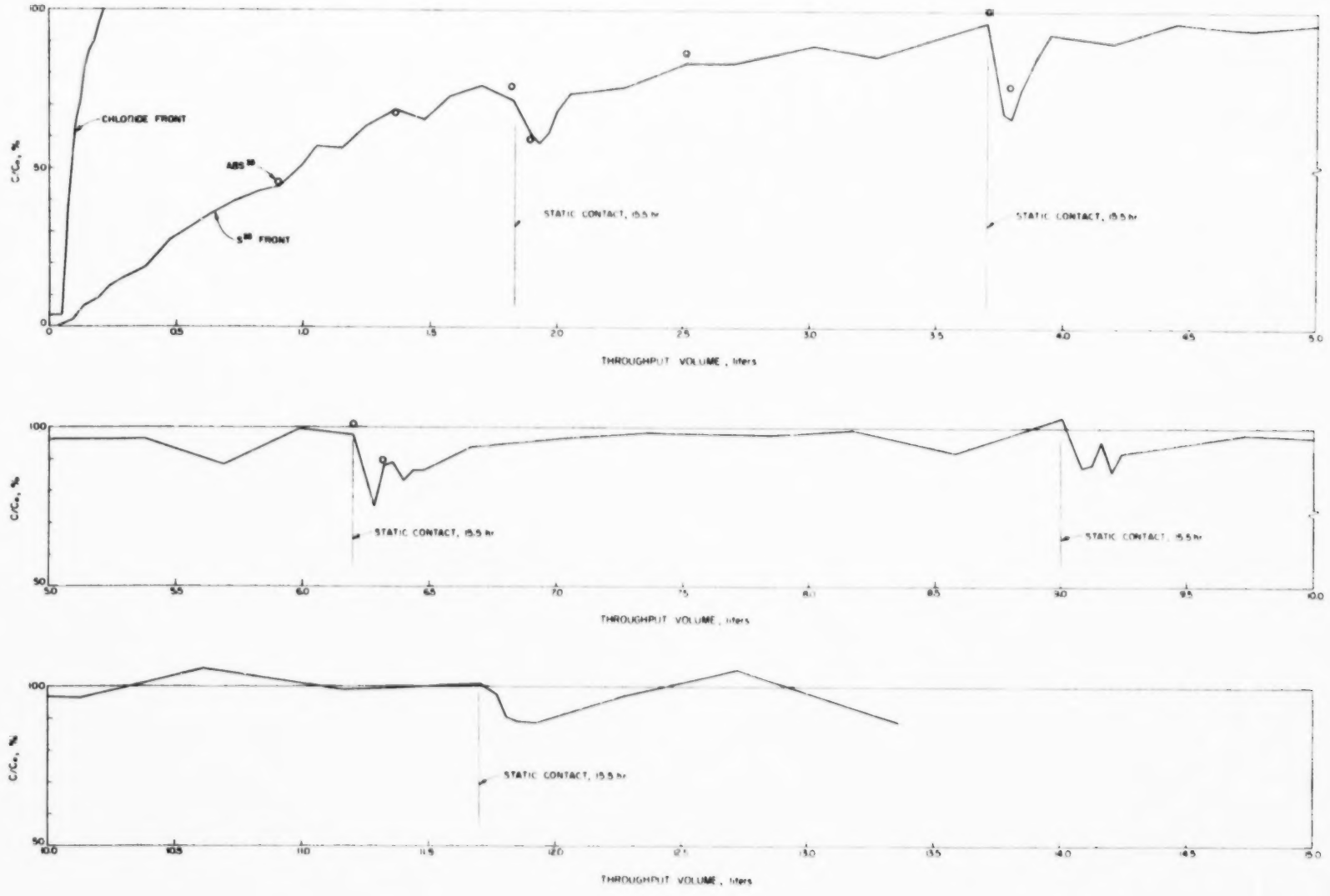


FIGURE 3 BREAKTHROUGH OF CHLORIDE, $S^{35}$, AND $ABS^{35}$ IN HANFORD (RUN 2) STERILE COLUMN

TABLE 3

ADSORPTION OF ABS ON FIVE CALIFORNIA SOILS*

| Throughput Volume, liter | ABS Adsorption, μg/g | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Oakley #3 | | | Yolo #2 | | | Hesperia | | | Hanford | | | Columbia | | |
| | Increment | % of Final Adsorption | Total | Increment | % of Final Adsorption | Total | Increment | % of Final Adsorption | Total | Increment | % of Final Adsorption | Total | Increment | % of Final Adsorption | Total |
| 1 | 1.14 | 33.6 | 1.14 | 1.94 | 33.4 | 1.94 | 2.23 | 42.3 | 2.23 | 2.85 | 40.0 | 2.85 | 2.40 | 32.2 | 2.40 |
| 2 | 0.23 | 6.8 | 1.37 | 1.16 | 20.0 | 3.00 | 0.17 | 32.2 | 3.40 | 1.50 | 21.1 | 4.35 | 1.12 | 15.2 | 3.52 |
| 3 | 0.03 | 0.9 | 1.40 | 0.55 | 9.5 | 3.55 | 0.42 | 8.0 | 3.82 | 0.84 | 11.8 | 5.19 | 0.81 | 10.9 | 4.33 |
| 4 | 0.14 | 4.1 | 1.54 | 0.25 | 4.3 | 3.80 | 0.35 | 6.6 | 4.17 | 0.55 | 7.7 | 5.74 | 0.60 | 8.0 | 4.93 |
| 5 | 0.21 | 6.2 | 1.75 | 0.53 | 9.1 | 4.33 | 0.37 | 7.0 | 4.54 | 0.26 | 3.7 | 6.00 | 0.47 | 6.3 | 5.40 |
| 6 | 0.17 | 5.0 | 1.92 | 0.04 | 0.7 | 4.37 | 0.40 | 7.6 | 4.94 | 0.25 | 3.5 | 6.25 | 0.30 | 4.0 | 5.70 |
| 7 | 0.05 | 1.5 | 1.97 | 0.14 | 2.4 | 4.51 | 0.27 | 5.1 | 5.21 | 0.34 | 4.8 | 6.59 | 0.20 | 2.7 | 5.90 |
| 8 | 0.23 | 6.8 | 2.20 | 0.34 | 5.9 | 4.85 | 0.07 | 1.3 | 5.28 | 0.08 | 1.1 | 6.67 | 0.19 | 2.5 | 6.09 |
| 9 | 0.39 | 11.5 | 2.59 | 0.29 | 5.0 | 5.14 | | | | 0.12 | 1.7 | 6.79 | 0.17 | 2.3 | 6.26 |
| 10 | 0.30 | 8.8 | 2.89 | 0.42 | 7.2 | 5.56 | | | | 0.23 | 3.2 | 7.02 | 0.54 | 7.2 | 6.80 |
| 11 | 0.23 | 6.8 | 3.12 | 0.24 | 4.1 | 5.80 | | | | 0.05 | 0.7 | 6.97 | 0.35 | 4.7 | 7.15 |
| 12 | 0.27 | 8.0 | 3.39 | | | | | | | 0.09 | 1.3 | 7.06 | 0.31 | 4.2 | 7.46 |
| 13 | | | | | | | | | | 0.05 | 0.7 | 7.11 | | | |

*Column conditions: sterile, saturated flow. Values are based on 1-mg/l ABS infiltrate.

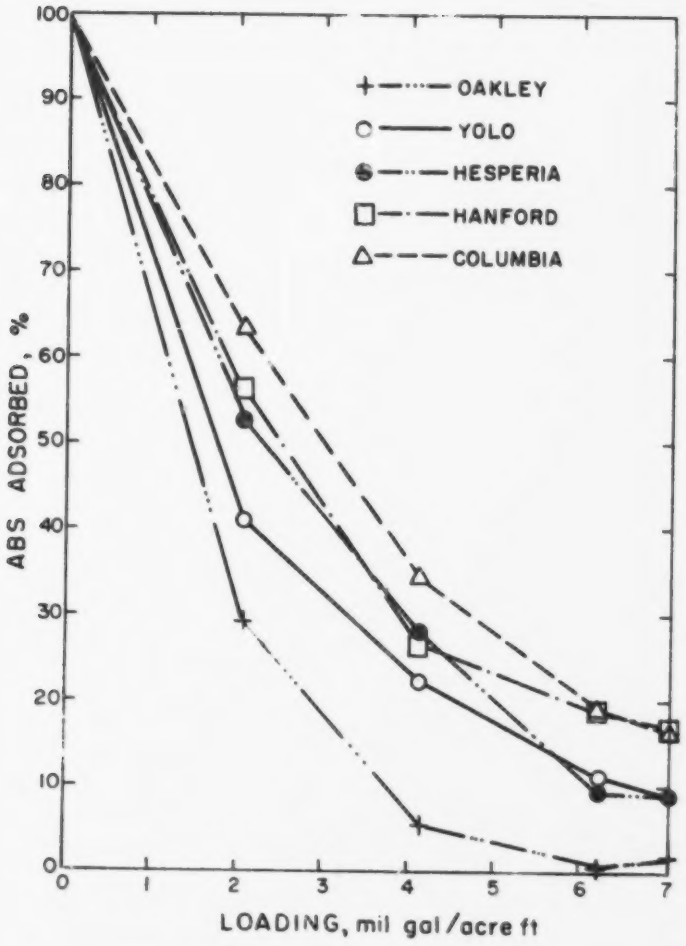


FIGURE 4. EFFECT OF INCREASED LOADING ON ABS REMOVAL BY FIVE SOILS INFILTRATED WITH 1ppm ABS CONCENTRATION (STERILE, SATURATED FLOW)

Nature of Adsorption of ABS

Analysis of the soil column after the end of percolation showed a consistent pattern of ABS adsorption. The greatest adsorption occurred at the top of the column, in the first 2.5 cm. Thereafter, the amount of ABS adsorption decreased to the bottom of the column (Figure 5). The course of ABS adsorption was found to follow the same general pattern illustrated in Figure 6. Four columns of Yolo sandy loam were infiltrated with ABS solution under sterile, saturated flow conditions for 1 hour, 3 hours, 7 hours, 16 hours, and 4 days, respectively (the last with three overnight static-contact periods). Flow rates were maintained as close as possible and feed solutions were of identical composition except for minute differences in ABS and $ABS^{35}$ concentrations. Figure 6 shows that the same pattern of ABS adsorption is followed in these columns as in the long-term column experiments. With two exceptions, there is a continuous reduction in ABS adsorption with soil depth (Table 4).

TABLE 4

BUILDUP AND DISTRIBUTION OF ADSORBED ABS ON STERILE YOLO SANDY LOAM WITH VARIABLE PERCOLATION TIME UNDER SATURATED FLOW

| Column | Zonal ABS Adsorption on Soil (μg/g) | | | | | |
|---|---|---|---|---|---|---|
| | 0 - 2.5 cm | 2.5 - 5.0 cm | 5.0 - 7.5 cm | 7.5 - 10 cm | 10 - 15 cm | 15 - 25 cm |
| 1-hour | 4.14 | 2.74 | 2.14 | 1.85 | 1.21 | 0.47 |
| 3-hour | 5.77 | 4.52 | 3.39 | 2.93 | 2.66 | 1.79 |
| 7-hour | 7.19 | 5.26 | 5.15 | 4.76 | 4.09 | 3.16 |
| 16-hour | 6.82 | 5.95 | 5.15 | 4.81 | 4.18 | 4.51 |
| 4-day* | 8.33 | 6.56 | 5.80 | 5.31 | 4.57 | 3.77 |

*Includes 3 static contact periods.

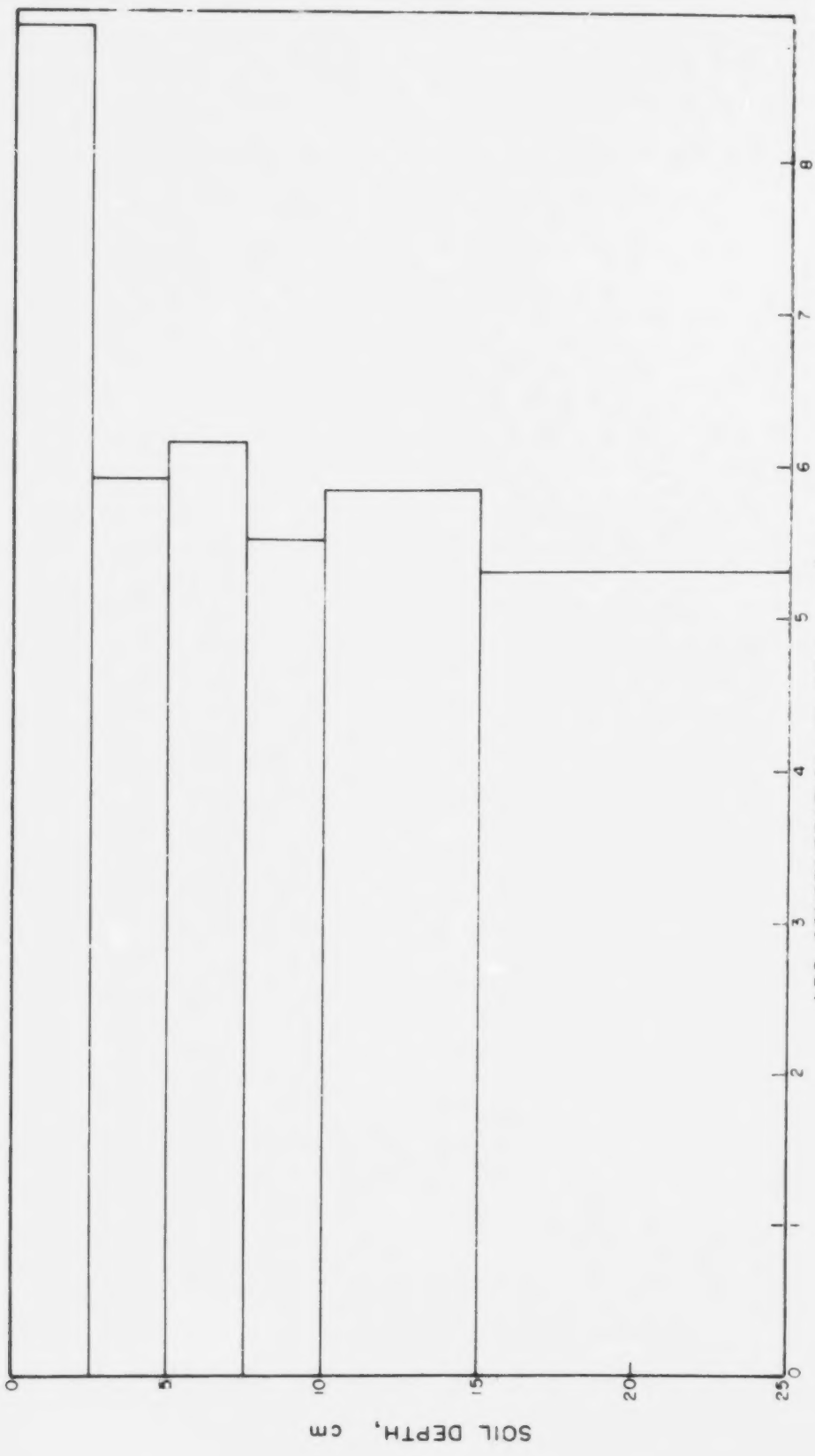


FIGURE 5. ZONAL ABS CONCENTRATIONS IN COLUMBIA SANDY LOAM AFTER PERCOLATION (STERILE, SATURATED FLOW)

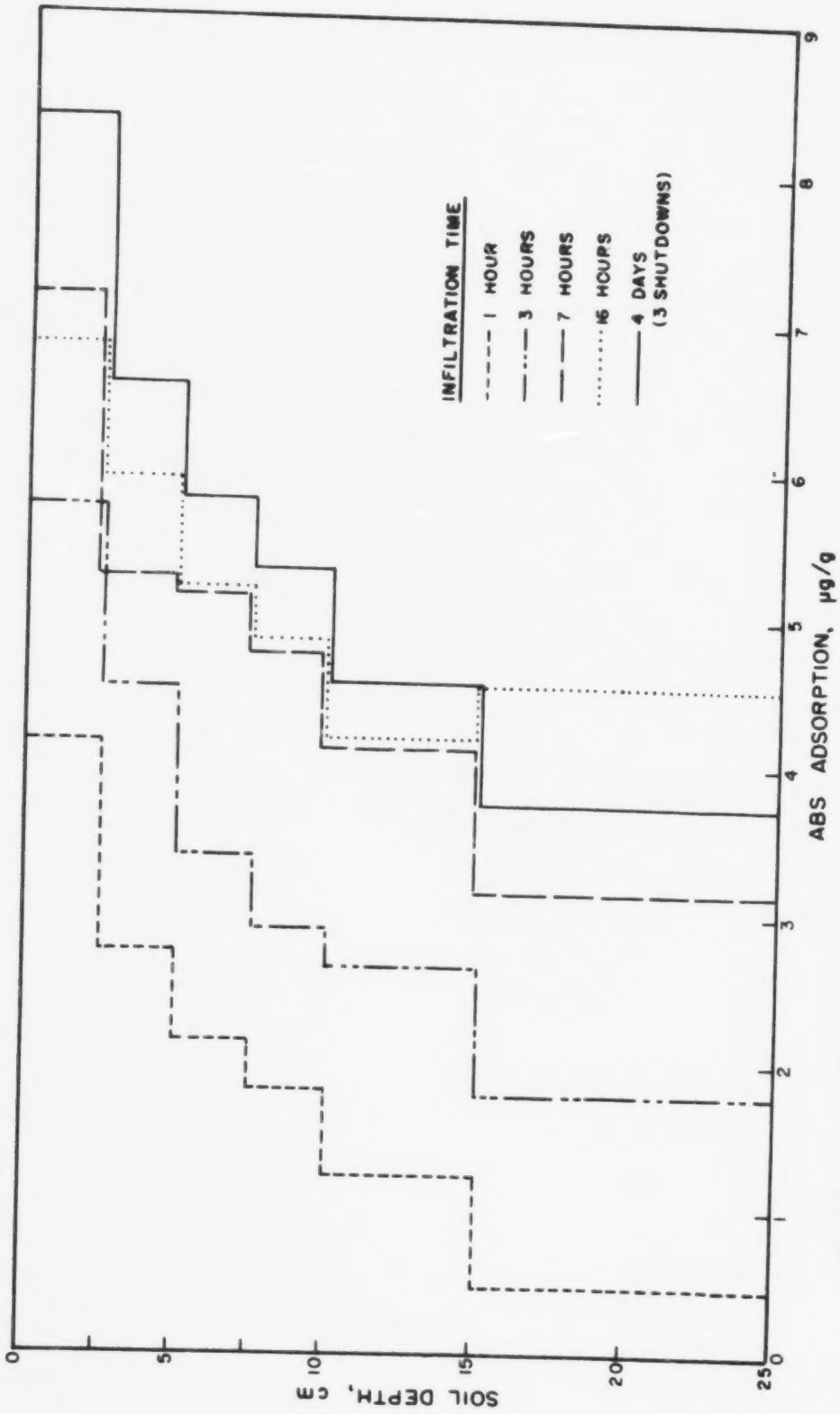


FIGURE 6. BUILDUP AND DISTRIBUTION OF ADSORBED ABS ON YOLO SANDY LOAM SUBJECTED TO VARIABLE PERIODS OF INFILTRATION (STERILE, SATURATED FLOW)

An adsorption isotherm of ABS on the bailings from a well-field aquifer located on the Richmond Field Station was constructed by shaking the bailings with solutions containing 10-, 5-, 0.1-, 0.01-, and 0.001-mg/l ABS and a minute amount of $ABS^{35}$. Analysis of the soil for adsorbed ABS gave results which fitted a Langmuir adsorption isotherm (Figure 7) where $M = KC^n$ when

K = constant,

M = amount of ABS adsorbed (mg/g),

and

C = concentration of ABS solution (mg/l).

By plotting log M and log C, a straight line was obtained where the slope n = 1. The value of K, the proportionality constant, was found to be $5.88 \times 10^{-4}$ l/g. In studies conducted at the University of Illinois (5), a K value of $5.96 \times 10^{-4}$ l/g was found for Ottawa sand in a concentration range of 1 to 100 mg/l, and a slope of 0.947. Adsorption is thus almost directly proportional to concentration up to 100 mg/l. A comparison of the ABS adsorption on well-field bailings by batch and column methods gave the extremely close results of $1.44 \times 10^{-3}$ mg/g for 1-mg/l ABS in the batch study and an average of $0.685 \times 10^{-3}$ mg/g for the column experiment.

The pattern of ABS adsorption noticed in column studies and the conformance of the batch experiments to the Langmuir adsorption isotherm strongly suggested that the adsorption of ABS conforms to the classical "plate theory" which postulates that an adsorbing medium removes an amount of solute from solution proportional to the concentration of that solution. If the medium were divided into a number of imaginary "plates," the solute adsorbed in each plate would be proportional to the concentration of its own infiltrate.

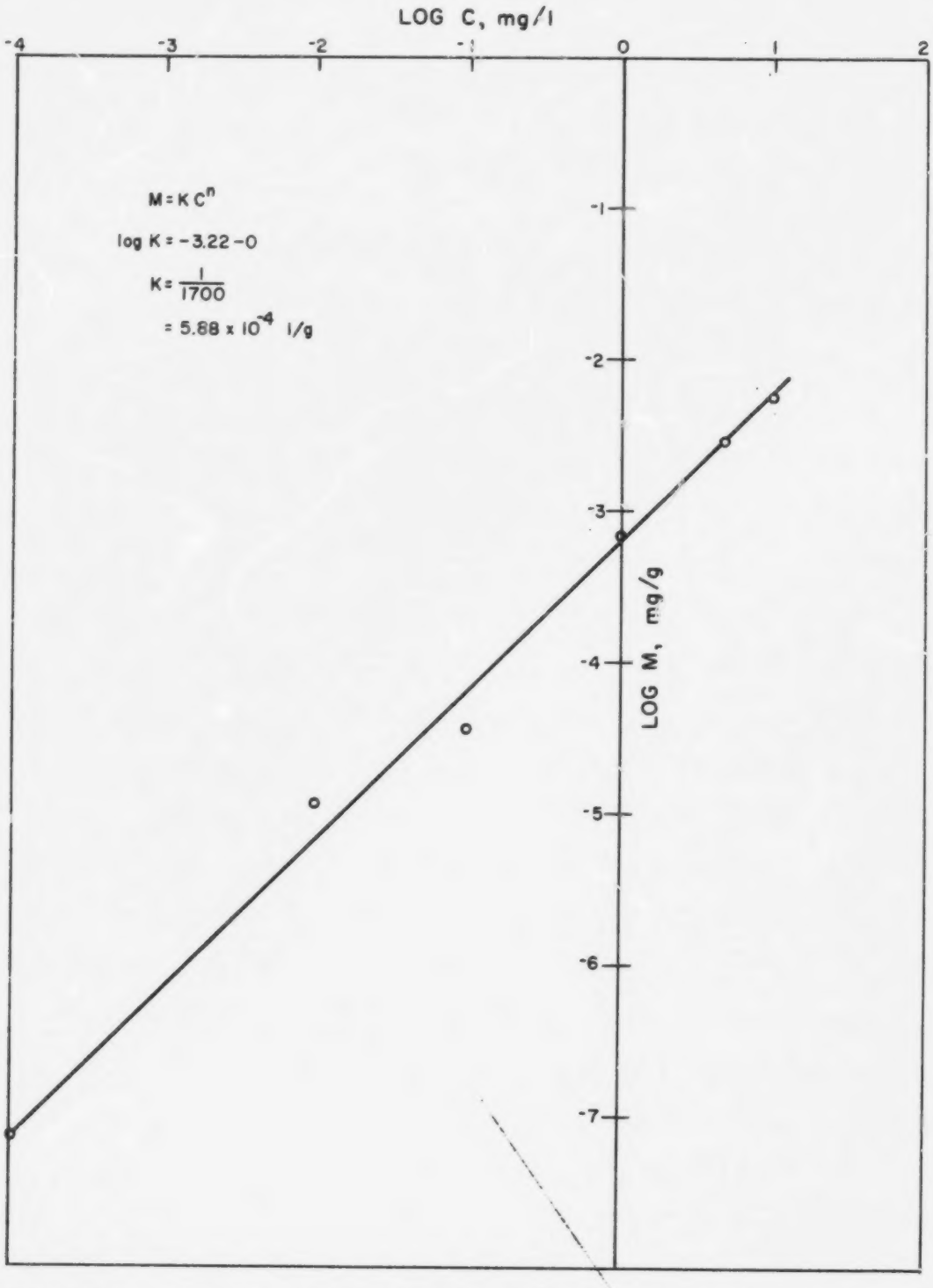


FIGURE 7. LOG OF ABS CONCENTRATION ON WELL-FIELD BAILINGS VERSUS LOG OF ABS IN LIQUID

Table 5 and Figure 8 show the high degree of conformance of experimental results to theoretical ABS adsorption values calculated on the basis of the plate theory. The slopes of the experimental lines in Figure 8 are almost identical to the theoretical curves except for the surface zone. Were the surface zone not considered, there would be far less displacement between actual and theoretical values. This is borne our further by Figure 9 which shows the near-perfect conformance of slopes based on the second zone for long-term runs on the five soils studied. It was concluded that ABS adsorption conforms to the plate theory except in the surface zone, where it is suspected that a molecular trapping of ABS molecules, as well as adsorption, takes place.

As would be expected from the previous experiments which show ABS adsorption on soils to be of the Langmuir type, it was found that the adsorption was rapidly reversible with ABS-free water to the extent of about 80 per cent of the ABS adsorbed.

Biologically Active Columns

The possibility that ABS removal by bio-degradation and other phenomena might occur during percolation of waste water through the biologically active mantle of the earth was investigated in a series of soil column studies. Should ABS removal in such a system prove significantly greater than in a sterile soil, the implication would be that surface spreading is a more suitable method of waste water reclamation by ground water recharge than is direct injection into underground strata. Percolation experiments were conducted on Oakley sand columns under both saturated and unsaturated soil conditions.

The two columns were seeded with sewage micro-organisms by percolating a mixture of 10 per cent sewage and nutrient broth equivalent to

TABLE 5

CONFORMANCE OF DISTRIBUTION OF ADSORBED ABS
ON YOLO SANDY LOAM TO PLATE THEORY

| Column | Initial Infiltrate ABS, µg | | Adsorbed ABS | | | |
|---|---|---|---|---|---|---|
| | | | µg | | Percentage* | |
| | Observed | Theory | Observed | Theory | Observed | Theory |
| DEPTH 0-2.5 cm | | | | | | |
| 1-hour | 413 | | 87.4 | | 21.2 | |
| 3-hour | 1,170 | | 116.7 | | 10.0 | |
| 7-hour | 2,919 | | 141.4 | | 4.8 | |
| 16-hour | 4,200 | | 140.2 | 140.2 | 3.3 | 3.3 |
| 4-day† | 6,562 | | 164 | 164 | 2.5 | 2.5 |
| DEPTH 2.5-5.0 cm | | | | | | |
| 1-hour | 325.6 | 325.6 | 57.9 | 68.6 | 14.0 | 16.6 |
| 3-hour | 1,053.3 | 1,053.3 | 91.4 | 105.3 | 7.8 | 9.0 |
| 7-hour | 2,777.6 | 2,777.6 | 103.4 | 133.0 | 3.5 | 4.6 |
| 16-hour | 4,059.8 | 4,059.8 | 122.3 | 132.8 | 2.9 | 3.2 |
| 4-day† | 6,398 | 6,398 | 129.1 | 160.2 | 2.0 | 2.4 |
| DEPTH 5.0-7.5 cm | | | | | | |
| 1-hour | 267.7 | 257.0 | 45.2 | 54.5 | 11 | 13.2 |
| 3-hour | 961.9 | 948.0 | 68.6 | 94.8 | 5.9 | 8.1 |
| 7-hour | 2,674.2 | 2,644.6 | 101.2 | 127.0 | 3.5 | 4.4 |
| 16-hour | 3,937.5 | 3,927 | 105.9 | 130.2 | 2.6 | 3.1 |
| 4-day† | 6,268.9 | 6,081.7 | 114.2 | 156.1 | 1.7 | 2.4 |
| DEPTH 7.5-10 cm | | | | | | |
| 1-hour | 222.5 | 202.5 | 39.1 | 43.0 | 9.5 | 10.4 |
| 3-hour | 893.3 | 853.2 | 59.3 | 85.4 | 5.1 | 7.3 |
| 7-hour | 2,573 | 2,517.6 | 93.7 | 121.1 | 3.2 | 4.1 |
| 16-hour | 3,831.6 | 3,796.8 | 99.0 | 126.0 | 2.4 | 3.0 |
| 4-day† | 6,154.7 | 5,929.4 | 104.5 | 152.3 | 1.6 | 2.3 |
| DEPTH 10-15 cm | | | | | | |
| 1-hour | 183.4 | 159.5 | 51.1 | 59.5 | 12.4 | 14.4 |
| 3-hour | 834 | 767.8 | 107.7 | 146.3 | 9.2 | 12.5 |
| 7-hour | 2,479.3 | 2,396.5 | 160.8 | 224.8 | 5.5 | 7.7 |
| 16-hour | 3,732.6 | 3,670.8 | 172.0 | 239.4 | 4.1 | 5.7 |
| 4-day† | 6,050.2 | 5,620 | 179.9 | 309.4 | 2.7 | 4.7 |
| DEPTH 15-25 cm | | | | | | |
| 1-hour | 132.3 | 100.0 | 39.6 | 61.5 | 9.6 | 14.9 |
| 3-hour | 726.3 | 621.5 | 145 | 214.1 | 12.4 | 18.3 |
| 7-hour | 2,318.5 | 2,171.7 | 248.8 | 388.2 | 8.5 | 13.2 |
| 16-hour | 3,560.6 | 3,431.4 | 370.8 | 428.4 | 8.8 | 10.2 |
| 4-day† | 5,870.3 | 5,100 | 296.8 | 520 | 4.5 | 7.9 |

*Percentage of initial infiltrate ABS adsorbed.

†Includes three contact periods.

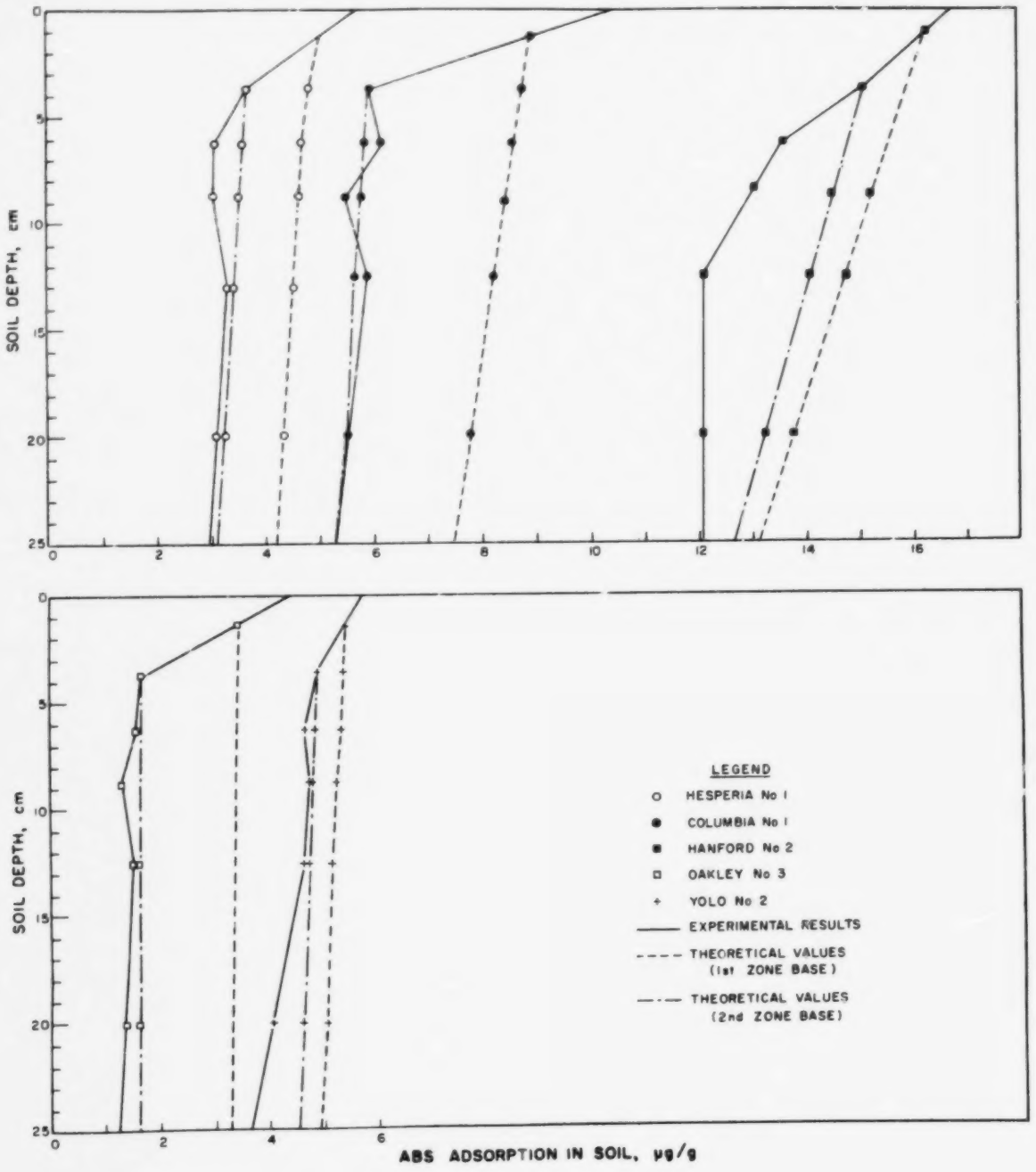


FIGURE 8. CONFORMANCE OF DISTRIBUTION OF ADSORBED ABS TO PLATE THEORY ON FIVE COLUMNS AFTER LONG-TERM DISCONTINUOUS INFILTRATION ON STERILE SOILS

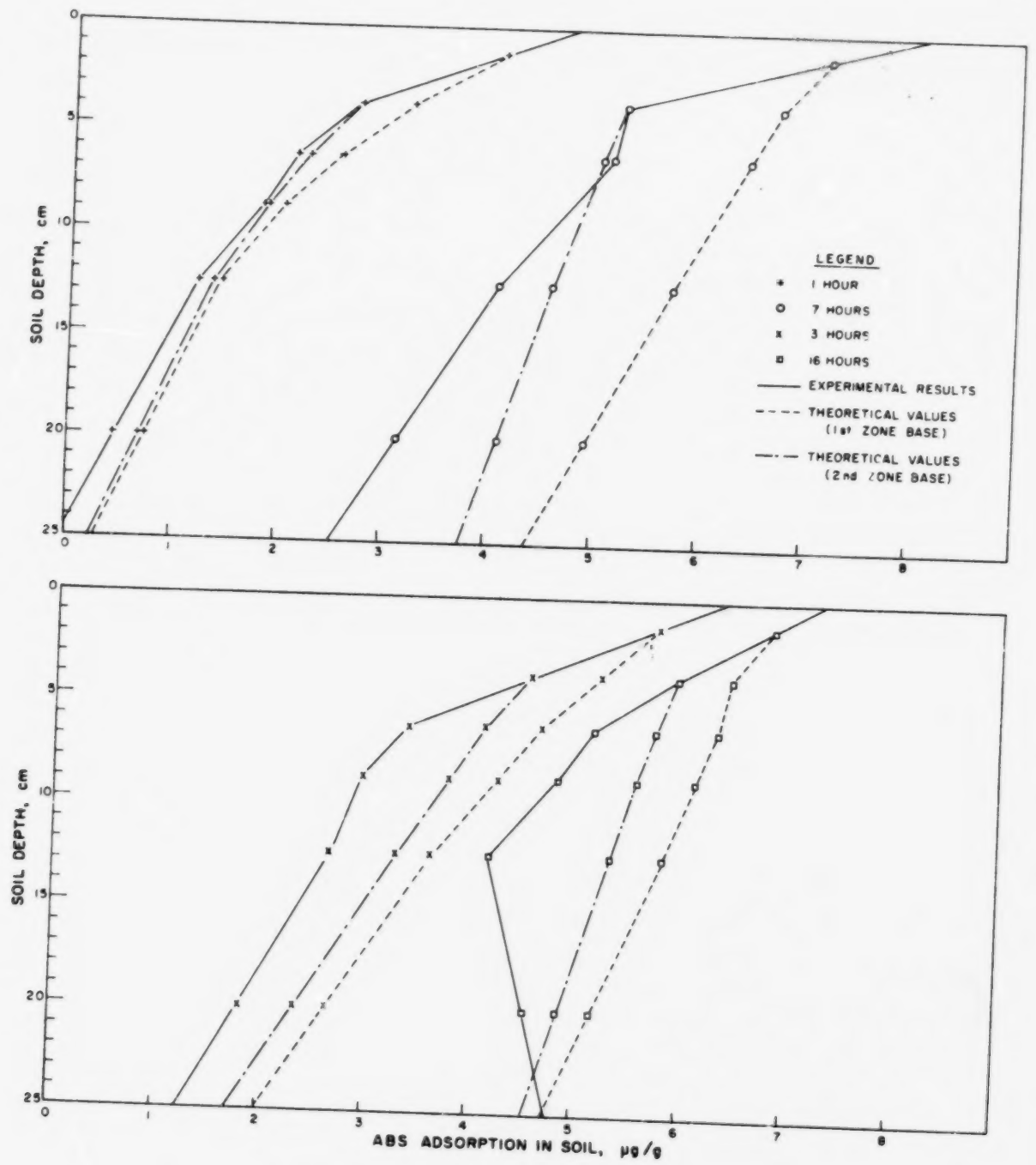


FIGURE 9. CONFORMANCE OF DISTRIBUTION OF ADSORBED ABS TO PLATE THEORY ON COLUMNS OF YOLO SANDY LOAM SUBJECTED TO VARIABLE PERIODS OF INFILTRATION (CONTINUOUS FLOW)

20 ppm BOD. The unsaturated column was drained completely before starting percolation of ABS solution while the saturated column was drained only to the surface of the soil. During the course of the experiment both columns were rested for periods varying between 16 and 358 hours. The saturated column was rested with a head of percolant maintained above it while the unsaturated column was allowed to drain and draw air into its pore spaces.

After these resting periods, the effluent from the two columns showed a distinctly different pattern of $S^{35}$ content. In the saturated column, the effluent $S^{35}$ showed the fall in concentration already noted in the sterile saturated columns after static contact periods. The $ABS^{35}$ and $S^{35}$ points corresponded very closely and at no time could any $S^{35}O_4$ be detected in the column effluent (Figure 10).

For the unsaturated column an initial total $S^{35}$ peak appeared in the effluent samples taken immediately after a static contact period (Figure 11). The corresponding $ABS^{35}$ values did not show such a pronounced peak and hence it must be concluded that some of the adsorbed $ABS^{35}$ was degraded to $S^{35}O_4$ and intermediate products which were then desorbed and washed out of the column by the first flush of percolant. That the $ABS^{35}$ points did show a considerable peak after the rest periods at 5.8 and 7.9 liters throughput can be attributed to evaporation, since during the following three periods, when evaporation was prevented by closing the top of the column, the $ABS^{35}$ points do not show any increase.

ABS degradation to intermediate products and $SO_4$ averaged 3.8 per cent subsequent to a complete activity breakthrough. The application rate was 2.5 ml per hour (0.12 million gal per acre per day) during this period.

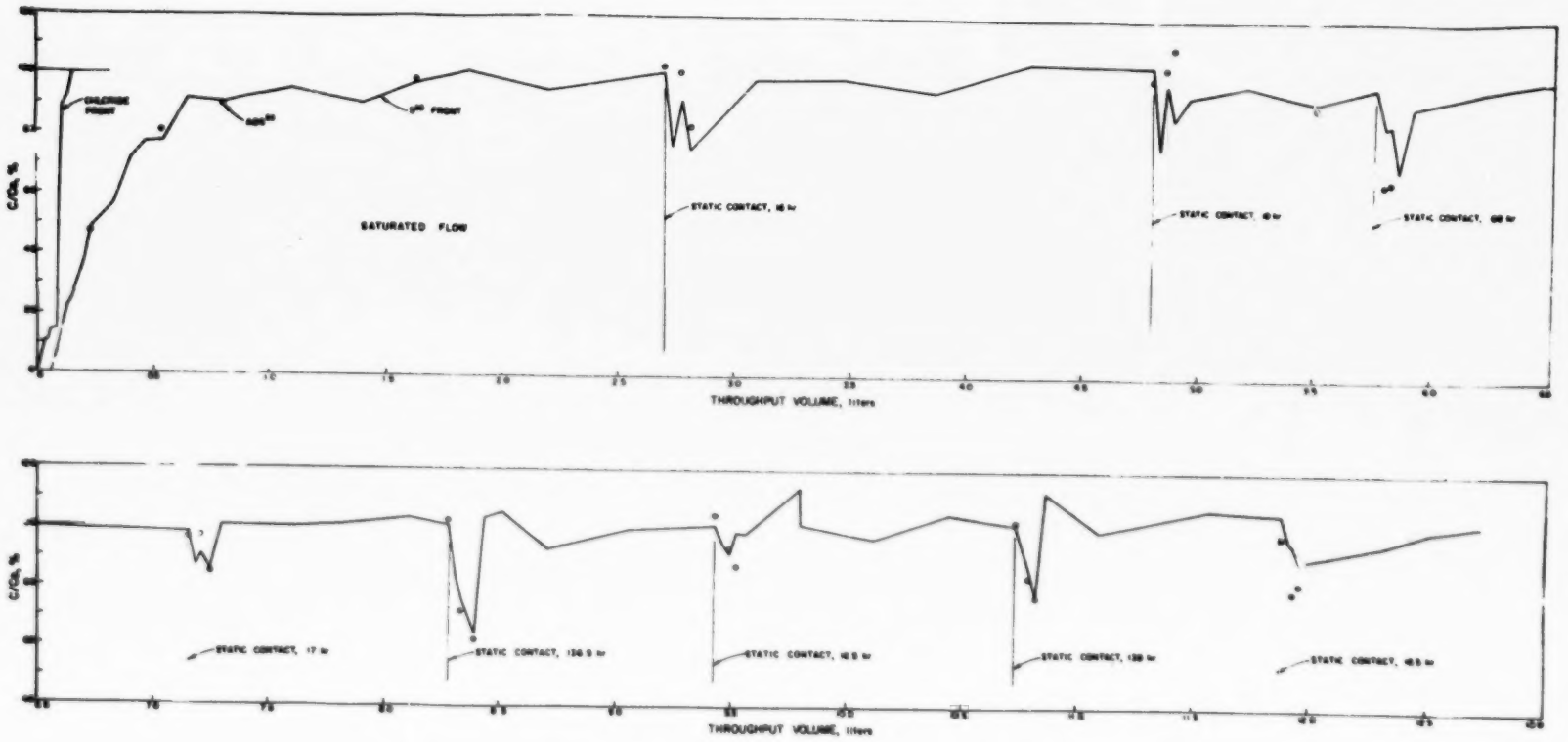


FIGURE 10. BREAKTHROUGH OF CHLORIDE, $S^{35}$, AND $ABS^{35}$ ON SATURATED, BIOLOGICALLY ACTIVE OAKLEY SAND

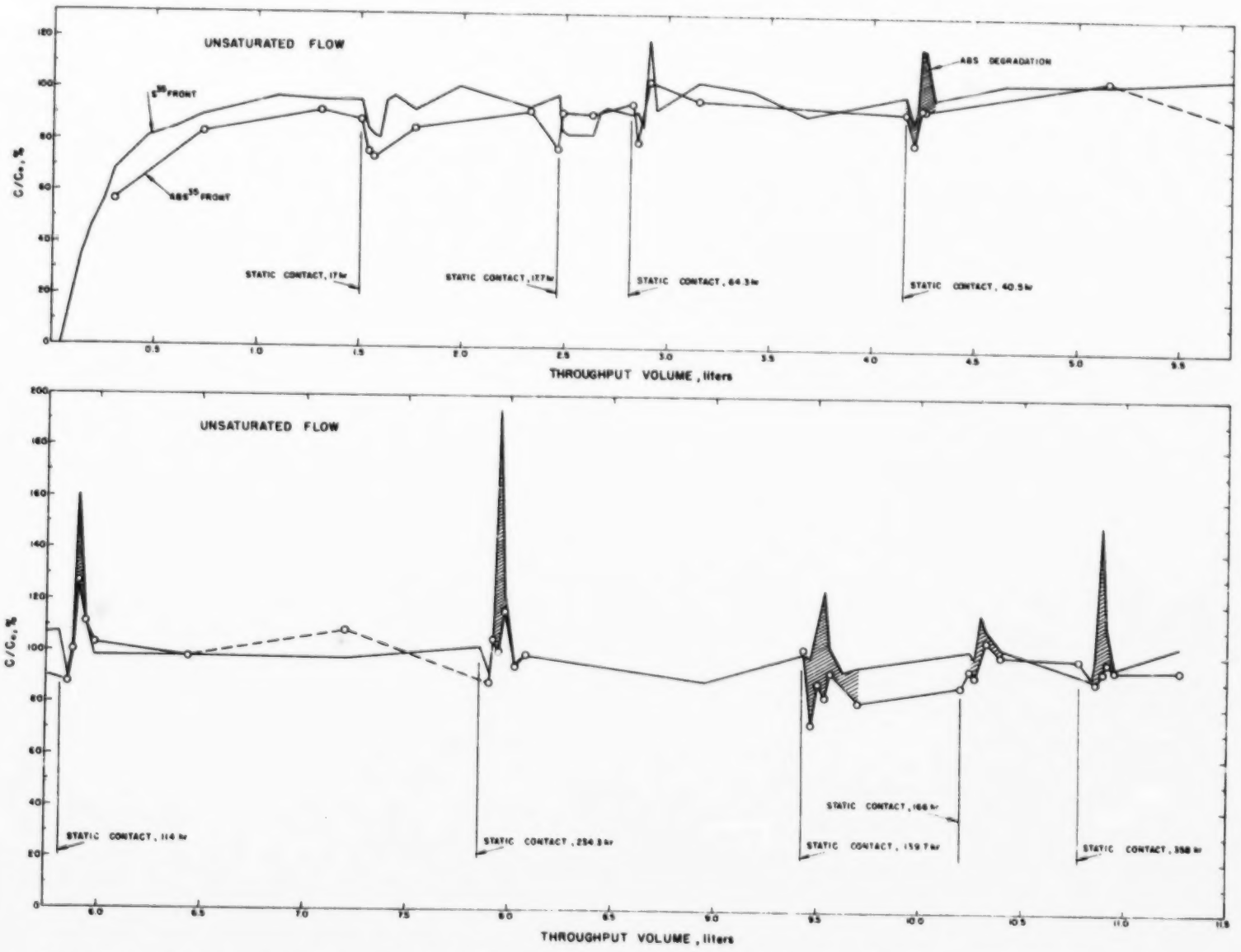


FIGURE II. BREAKTHROUGH CURVES OF $S^{35}$ AND $ABS^{35}$ ON UNSATURATED, BIOLOGICALLY ACTIVE OAKLEY SAND, SHOWING $ABS^{35}$ DEGRADATION

The rate of ABS degradation to $SO_4$, computed for five of the rest periods, is given in Table 6. This rate averaged 0.0106 µg per hour of static contact time or an insignificant 0.003 per cent of the adsorbed ABS. For a soil of greater ABS adsorptive capacity than Oakley sand, such as Hanford, this rate would be only 0.0002 per cent of the adsorbed ABS.

TABLE 6

RATE OF DEGRADATION OF ABS BY BIOLOGICALLY ACTIVE OAKLEY SAND

| Shutdown Time, hr | ABS Degradation to $SO_4$ | | |
|---|---|---|---|
| | Quantity, µg | Rate | |
| | | µg/hr | mg/hr/acre-ft |
| 40.5 | 0.305 | 0.0075 | 37 |
| 234.3 | 6.243 | 0.0266 | 131 |
| 139.7 | 3.225 | 0.0231 | 114 |
| 166.0 | 2.846 | 0.0171 | 84 |
| 358.0 | 11.491 | 0.0321 | 158 |
| Average | | 0.0106 | 105 |

Although only one soil was examined and at but one concentration of infiltrate ABS, it is probable that no significant increases in degradation would occur in other soils or at other concentrations. This is presumed from experiments in which increasing doses of ABS to activated sludge units caused no increase in ABS degradation (3). If concentration had no effect under the highly favorable oxidation conditions of activated sludge treatment, then it is almost certain that this would hold true for the quite limited oxidative processes in soils. The same reasoning would apply to different soil types.

It seems evident that, except for the surface zone, there is, for practical purposes, little difference in ABS adsorption between biologically active and inactive soils, regardless of condition of air-binding or saturated flow.

Degradation occurs only in biologically active, air-bound soil, and predominantly in the surface zone.

An effort was made to increase the amount of ABS degradation in a biologically active, unsaturated aerobic soil column. A column of Hanford sandy loam was seeded with glass-wool filtered settled sewage for a period of nine days. Between each day's dose of sewage, the soil column was drained by applying suction to the effluent tube.

After 100 per cent breakthrough of $S^{35}$ and $ABS^{35}$ was obtained, the column was operated on a "percolation with static contact" schedule. During this period the average loading rate was 1.1 mil gals/acre/day, inclusive of static contact periods. The average ABS breakdown to $SO_4$ and intermediate products was 25 per cent.

When a dosing syphon was installed to deliver 35 ml feed to the column every six hours (an over-all loading rate of 0.29 mil gals/acre/day), the average ABS breakdown increased to 35 per cent. Thus a decrease in over-all loading rate of about 72 per cent produced an increase in ABS breakdown of only 10 per cent.

An investigation was made to determine the effect on ABS degradation of a coarse soil overlaid by a strata of fine soil to simulate natural soil stratification as it frequently exists in the field. The limitation on percolation rate imposed by the fine soil prevents saturation of the lower soil strata during continuous percolation and therefore represents an ideal natural situation for ABS degradation.

The experimental apparatus was two 2.5-cm diameter columns consisting of a 14-cm long column of Hanford Fine Sandy Loam positioned above and connected by rubber tubing to a 94-cm long column of Oakley Sand.

A ten per cent sewage solution was percolated through the columns for one week. Following this, 6.7 liters of sterile simulated sewage effluent containing 1-mg/l ABS, tracer $ABS^{35}$, and approximately 20-mg/l BOD was percolated through the columns over a twenty-four-day period. It was not possible to maintain sterile conditions in the feed solution, so in order to retard ABS breakdown in the feed reservoir, a new solution was prepared which contained no nutrient broth or phosphate. Over a seventeen-day period, 5.4 liters of this solution was percolated continuously through the column. Throughout the experiment the liquid in the column of Hanford soil was aerated to insure aerobic conditions.

During the last three days of the experiment, the total activity breakthrough averaged 91.2 per cent and the ABS breakthrough averaged 70.7 per cent. Thus ABS degradation--the difference between these two figures--was 20.5 per cent. Complete degradation to inorganic sulfur during this period averaged 2.3 per cent. The loading rate was 0.65 mil gal/acre/day compared to 1.1 mil gal/acre/day for the previous experiment where the ABS degradation was 25 per cent. Thus a 38.5 per cent decrease in loading rate resulted in a 4.5 per cent decrease in ABS degradation in this experiment.

## SUMMARY AND CONCLUSIONS

Experiments were run on the adsorption and bio-degradation of ABS from water percolating through each of five soil types under saturated and

unsaturated, and sterile and biologically-active conditions, and under continuous and intermittent flow conditions. The results of these experiments support the following conclusions:

1. All soils examined displayed an ability to adsorb ABS (1.5 to 10 μg/g for an infiltrate containing 1-mg/l ABS).
2. The degree of adsorption was not sufficient to prevent an early breakthrough of ABS into ground water. At a loading of 6 million gallon/acre foot the breakthrough of ABS is 98.5% for Oakley, 88.5 for Hesperia, 87.5 for Yolo, 81.0 for Hanford, and 79.0 for Columbia.
3. Adsorption isotherms for ABS are linear up to 100 mg/l.
4. ABS adsorption conforms to the classical "plate theory" which states that an adsorbing medium will adsorb a solute in proportion to the solute concentration. Therefore, as ABS is adsorbed from an infiltrate passing through soil, there is a continuous decrease in ABS adsorption.
5. ABS is rapidly desorbed from soil to an extent of approximately 80 per cent when flushed with ABS-free water.
6. Under saturated flow conditions no ABS degradation occurs.
7. There is no difference in ABS adsorption between saturated and unsaturated flow conditions.
8. ABS degradation occurs only in biologically active, air-bound soil and predominantly in its upper 2.5 cm.
9. Maximum ABS degradation occurs when percolation is intermittent (at a loading rate of 1.1 million gallons per acre per day, ABS degradation averaged 25 per cent; at 0.29 million gallons per acre per day, it averaged 35 per cent).

10. Small doses applied at frequent, regular intervals enhance ABS degradation in contrast to large doses applied infrequently (in the latter case only 3.8 per cent ABS degradation occurred at a loading rate of 0.12 million gallons per acre per day).
11. Continuous percolation through a coarse soil overlaid by a finer soil gives substantial ABS degradation (20.5 per cent at a loading rate of 0.65 million gallons per acre per day).
12. ABS degradation in biologically active soil is insufficient to prevent contamination of ground water.

## Part II. EFFECT ON PLANT GROWTH

### INTRODUCTION

Direct reuse of reclaimed waste waters in irrigated agriculture in semi-arid areas is becoming an increasingly attractive possibility. In some places it is already an accomplished fact. Substitution of sewage or sewage effluents for the normal sources of irrigation water, however, poses the question of whether the value of the water in sewage and sewage effluents is in any way decreased by its contaminants. That it has value as a fertilizer was realized and practiced by the Ancient Chinese, but today, with its content of chemically synthesized substances, this value may be somewhat offset. A grower would probably prefer to pay more for normal irrigation water than to use a substitute which would reduce his crop even by a few per cent. The persistence of ABS during sewage treatment therefore gives a degree of urgency to studies on the effects of the present type of detergents upon plant growth, and of the uptake of detergents by plants (especially the edible portions). Such studies are reported in this paper.

Experiments on ABS-uptake and movement in plants were first carried out using the water culture method (1) which is commonly employed in toxological studies because of rapidity of plant growth. Subsequently, the experiments were repeated with plants rooted in normal agricultural soil. Two plant species, the dicotyledon (sunflower) and the monocotyledon (barley), were chosen for these studies because of their rapid growth and resistance to insects and disease. Included in soil studies was a third species, Lupinus albus (a dicotyledon).

## PLANT GROWTH IN WATER

### Apparatus and Methods

The apparatus consisted of nine 20-liter containers fitted with air lines to provide oxygen to the roots. Seedlings, germinated and grown in vermiculite for approximately one week, were supported over the containers in corks fitted in a wooden platform. A dacron-wool wrapping at the base of the stems secured the seedlings in the corks and yet provided sufficient flexibility to accommodate plant growth. Light was provided by a bank of fluorescent lights which were turned on for 16 hours each day by an automatic timer.

Five plants were grown in each container and there were three containers for each of the three ABS concentrations tested. The "magic square" method of randomization was used to determine the position of containers under the light bank.

Tracer $ABS^{35}$ was added to the water culture along with the carrier ABS, in order that a measurement of the plant's uptake of ABS could be made by radioassay. Foaming was controlled by the addition of a foam depressant to each container.

### Sunflower Experiments

In the first experiment sunflowers were grown in nutrient solutions containing 0, 10, and 40 mg/l ABS, respectively, for a period of twelve days. Growth was estimated by measuring leaf length and epicotyl length at intervals throughout the experiment. At the end of the growth period, the roots and stems were separated. Special care was taken to wash off the nutrient solution from the roots with distilled water. The roots and stems were weighed, oven-dried at 105°C, reweighed, and ground to a fine powder.

Effect of ABS on Growth

After twelve days growth, the average length of the first leaves of the control plants was almost double that of the 10-mg/l plants, and 4.5 times the 40-mg/l plants (Figure 12). Similar inhibition of epicotyl length is shown in Figure 13. Photographs taken after 12 days of growth (Figures 14, 15, and 16) illustrate the inhibition produced by ABS. In these photographs it is notable that the control plants have lush roots and foliage while the plants grown in 40-mg/l ABS appear particularly retarded, and even at 10-mg/l ABS a marked growth inhibition appears. The extent of inhibition is shown by final plant weights (Table 7). The fresh weight of the control plants was 12.5 times that of the 40-mg/l plants, and about 4 times that of the 10-mg/l plants. The percentage of water content was also higher in the control plants.

TABLE 7

EFFECT OF ABS ON GROWTH OF SUNFLOWER PLANTS

| ABS, mg/l | Mean Weight, g | | Water Content, Fresh Weight Basis, % |
|---|---|---|---|
| | Fresh | Dry | |
| 0 | 8.22 | 0.72 | 91.2 |
| 10 | 2.12 | 0.22 | 89.8 |
| 40 | 0.65 | 0.09 | 86.2 |

Not only were growth differences apparent, but, as may be seen in Figure 17, the leaves of the 10-mg/l plants were extremely chlorotic. A yellowing of new leaves and of older leaves near the stem indicated that even at fairly low concentrations, ABS can cause chlorosis.

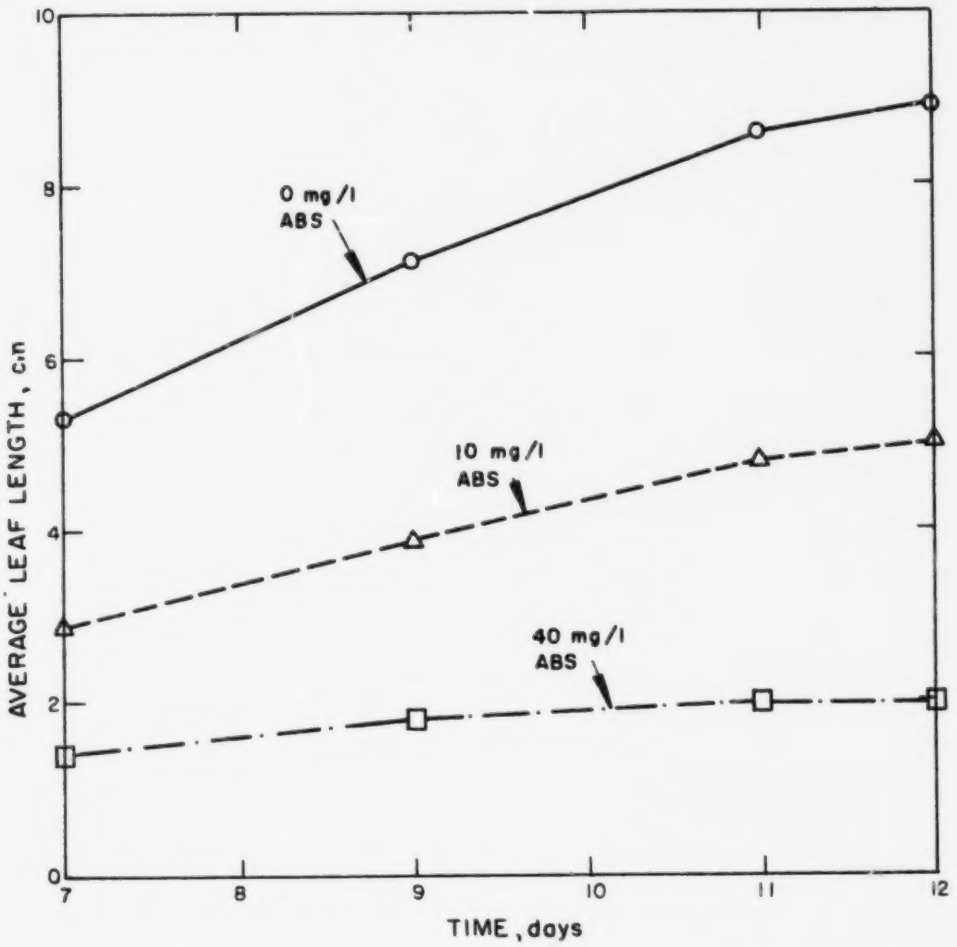


FIGURE 12. GROWTH OF SUNFLOWERS IN WATER CULTURE, LEAF MEASUREMENTS

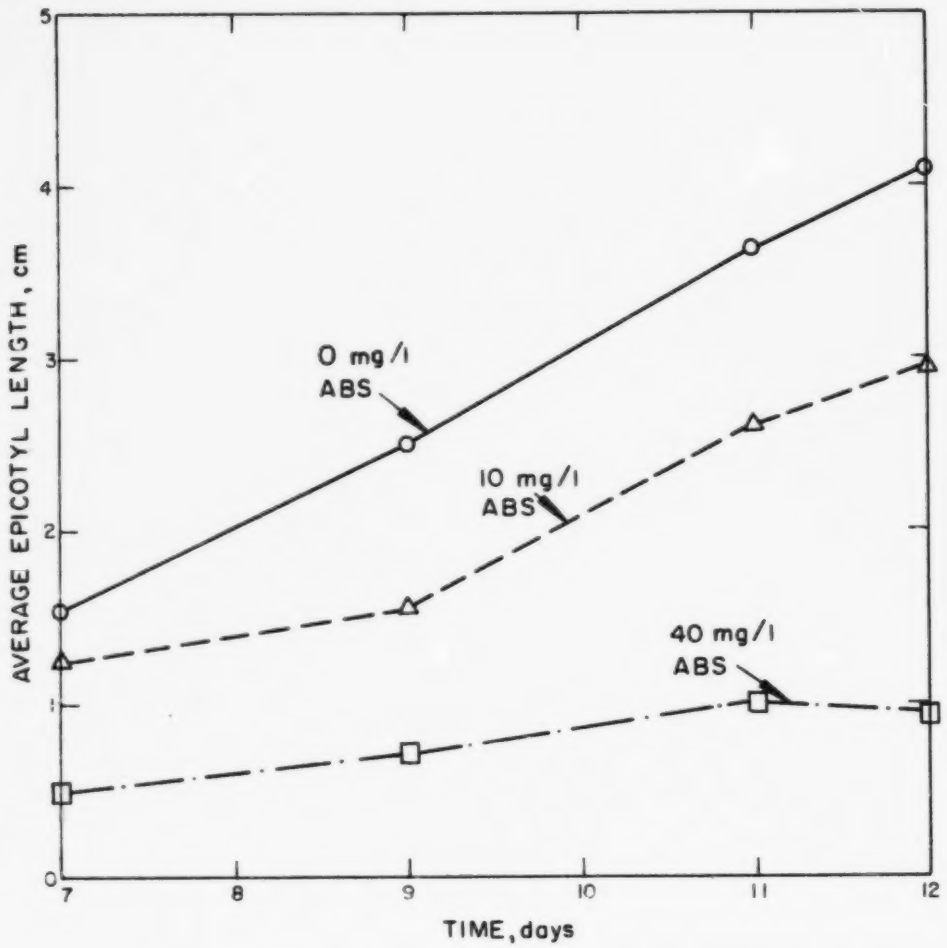


FIGURE 13. GROWTH OF SUNFLOWERS IN WATER CULTURE, EPICOTYL MEASUREMENTS

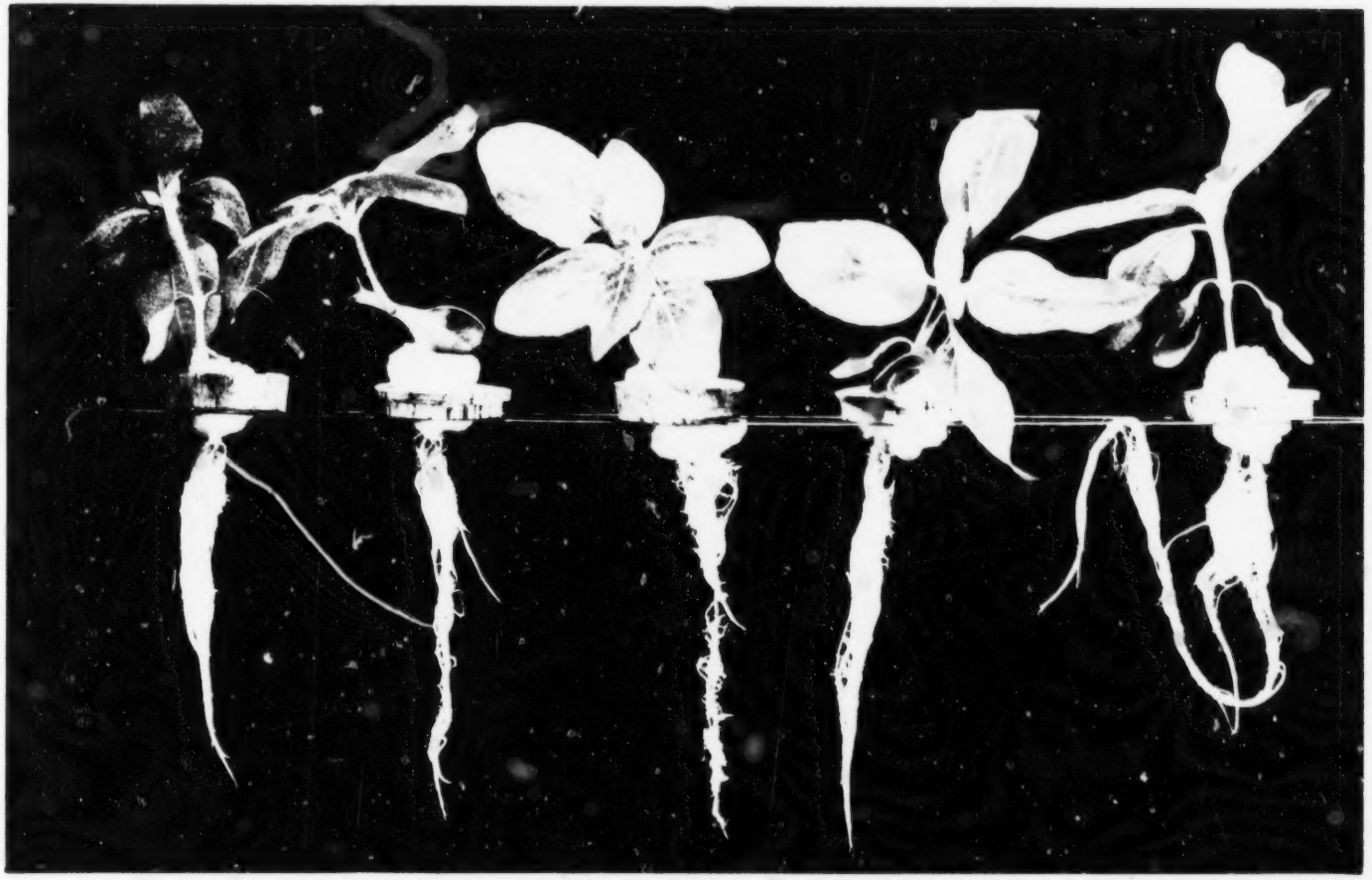

FIGURE 14. CONTROL SUNFLOWERS GROWN IN WATER CULTURE.

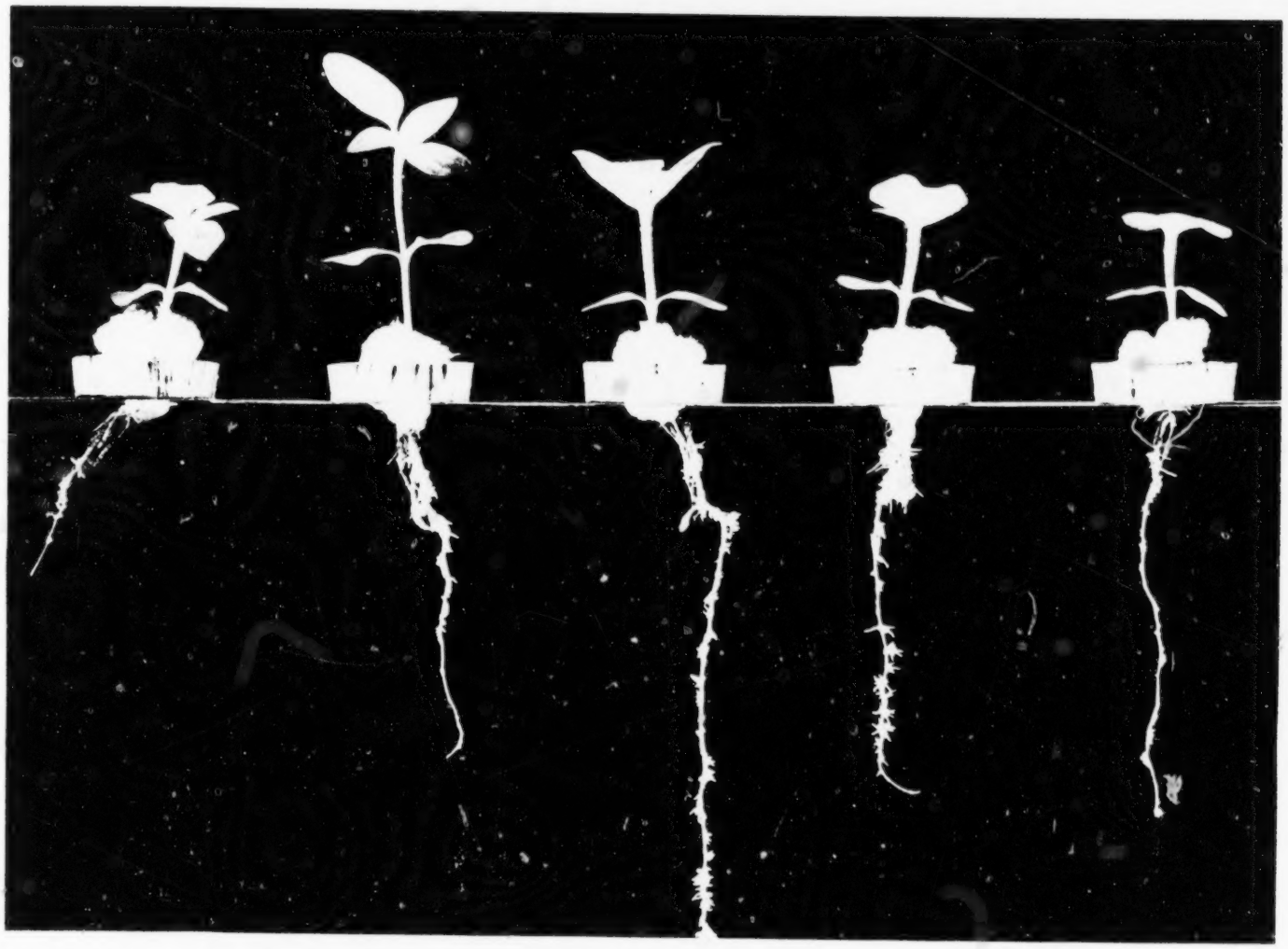

FIGURE 15.—SUNFLOWERS GROWN IN 10 MG. L ABA WATER CULTURE.

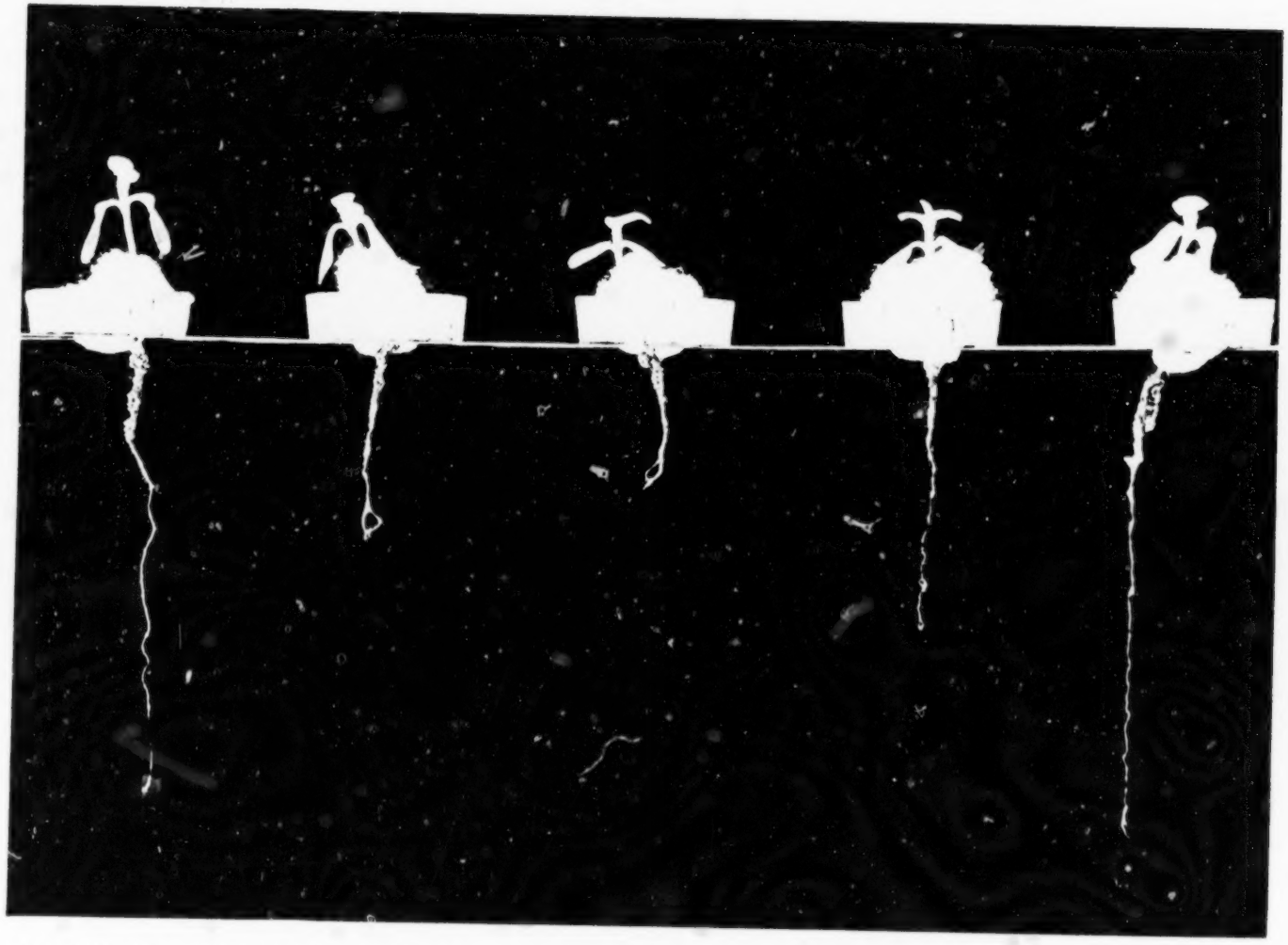

FIGURE 16.—SUNFLOWERS GROWN IN 30 MG. L. ABS WATER CULTURE.

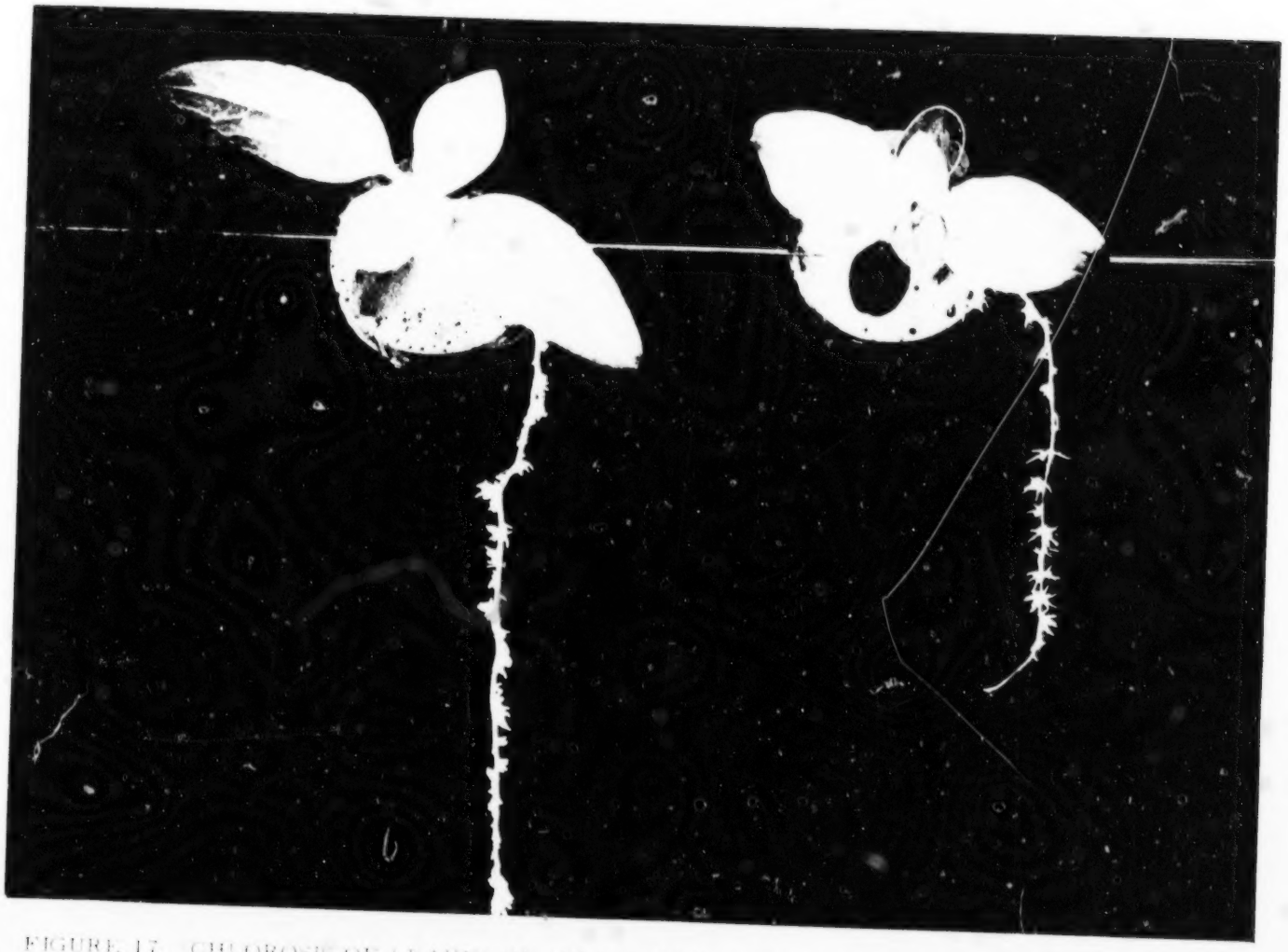

FIGURE 17. CHLOROSIS OF LEAVES OF SUNFLOWERS GROWN IN 10 MG. L. ABS. WATER CULTURE.

Uptake of ABS

Radioassay of the test plants revealed that ABS is concentrated in the roots and is also present, but to a lesser extent, in the stems and leaves. The ABS values, shown in Table 8, are obtained by conversion of the measured $ABS^{35}$ to total ABS using the known tracer-to-carrier ratio of the liquid. The S values are derived from the measured $S^{35}$, or total activity, and converted in the same manner. S values thus refer to all sulfur products derived from the carrier and tracer, and therefore include degradation products of ABS as well as any unaltered ABS. Plant uptake of ABS increased with increasing ABS concentration in the water culture. This increase is not proportional, however, since a 4-fold increase, from 10 to 40 mg/l, in liquid ABS produced a 22-fold increase in root and a 37-fold increase in stem and leaf ABS concentration.

TABLE 8

AVERAGE UPTAKE OF ABS AND S BY SUNFLOWERS GROWN IN WATER CULTURE

| Water Culture ABS, mg/l | Plant Part | S, µg/g dry weight | ABS, µg/g dry weight | Degradation of ABS, % |
|---|---|---|---|---|
| 10 | Stems and leaves | 154 | 87 | 43.5 |
| 10 | Roots | 4,740 | 4,300 | 9.3 |
| 40 | Stems and leaves | 4,123 | 2,377 | 42.4 |
| 40 | Roots | 93,100 | 93,100 | 0 |

Degradation products of ABS were found predominantly in the stems and leaves, which would seem to indicate that this is where degradation occurs. However, it is possible that degradation actually occurs in the roots, and that degradation products are then rapidly translocated to the stems and leaves.

Barley Experiments

The experimental procedure for growing barley was similar to that for sunflowers. Atlas barley was germinated in vermiculite and then grown for 14 days in water culture containing ABS concentrations of 0, 10, and 40 mg/l. Tracer $ABS^{35}$ was added, not only to the test containers, but also to one of the controls. No foam depressant was used in this experiment to eliminate the remote possibility of a synergistic effect with ABS on the test plants, which would not occur in the ABS-free control.

The apparatus was altered slightly to give needed support to the barley seedlings. Individual supports were constructed using thread wound around two upright glass rods.

Plant growth was noted during the course of the experiment by measurement of shoot length. Each new shoot was measured from the ligule of the old shoot.

Results showed that barley was inhibited by ABS to the same extent as sunflowers. Figure 18 depicts the average growth of the second shoots which is typical of over-all plant growth. Again there was virtually no growth at 40-mg/l ABS, and, as shown in Table 9, the mean fresh weights of the controls were approximately three times that of the 10-mg/l plants. The appearance of the plants (Figure 19) shows the adverse effects at the higher ABS concentration.

TABLE 9

EFFECT OF ABS ON GROWTH OF BARLEY PLANTS IN WATER CULTURE

| ABS, mg/l | Mean Weight, g | | Mean Water Content, Fresh Weight Basis, % |
|---|---|---|---|
| | Fresh | Dry | |
| 0 | 1.49 | 0.17 | 88.5 |
| 10 | 0.51 | 0.09 | 82.5 |
| 40.0 | 0.21 | 0.06 | 71.4 |

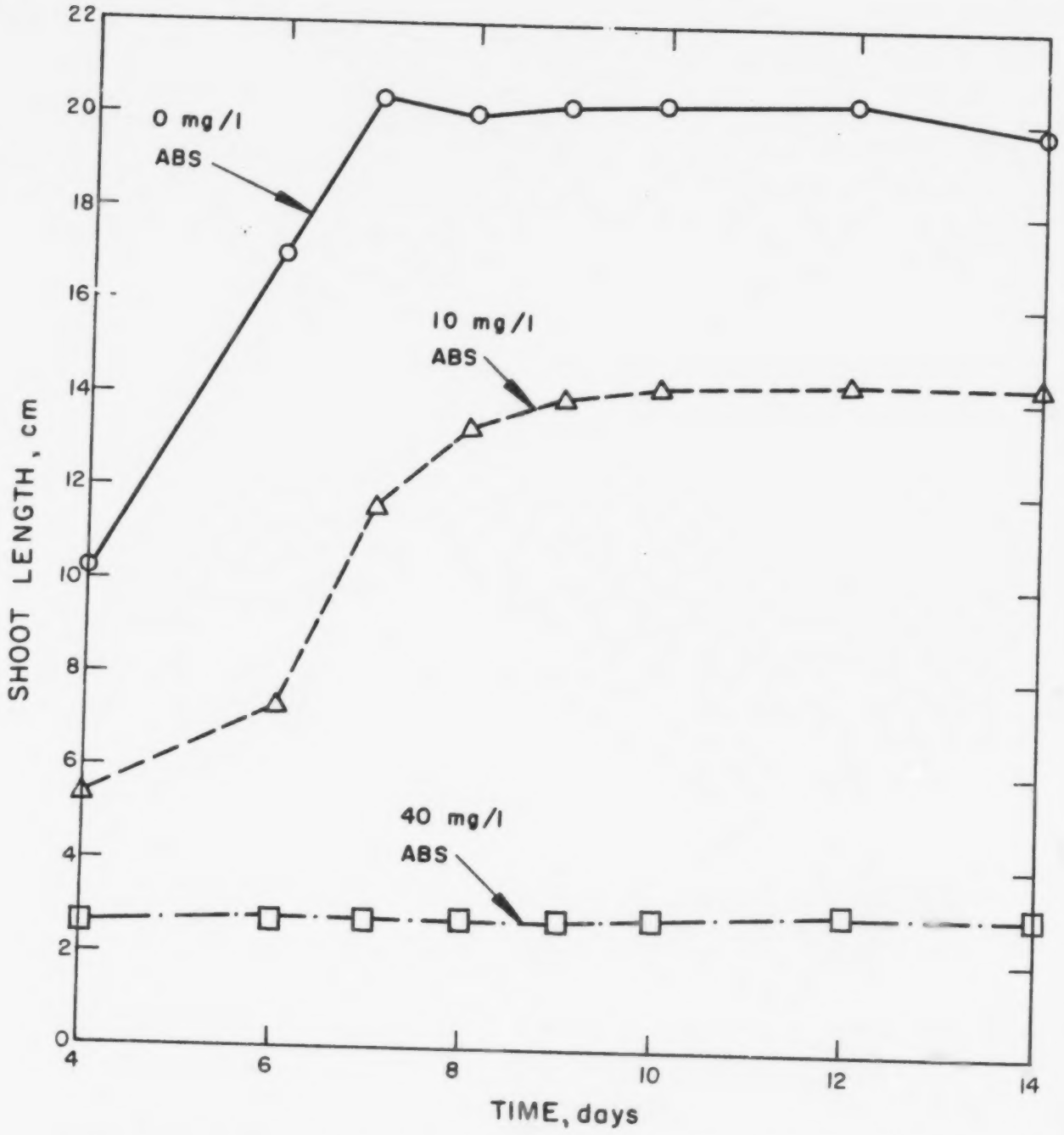


FIGURE 18. GROWTH OF BARLEY ( 2nd Shoot ) IN WATER CULTURE

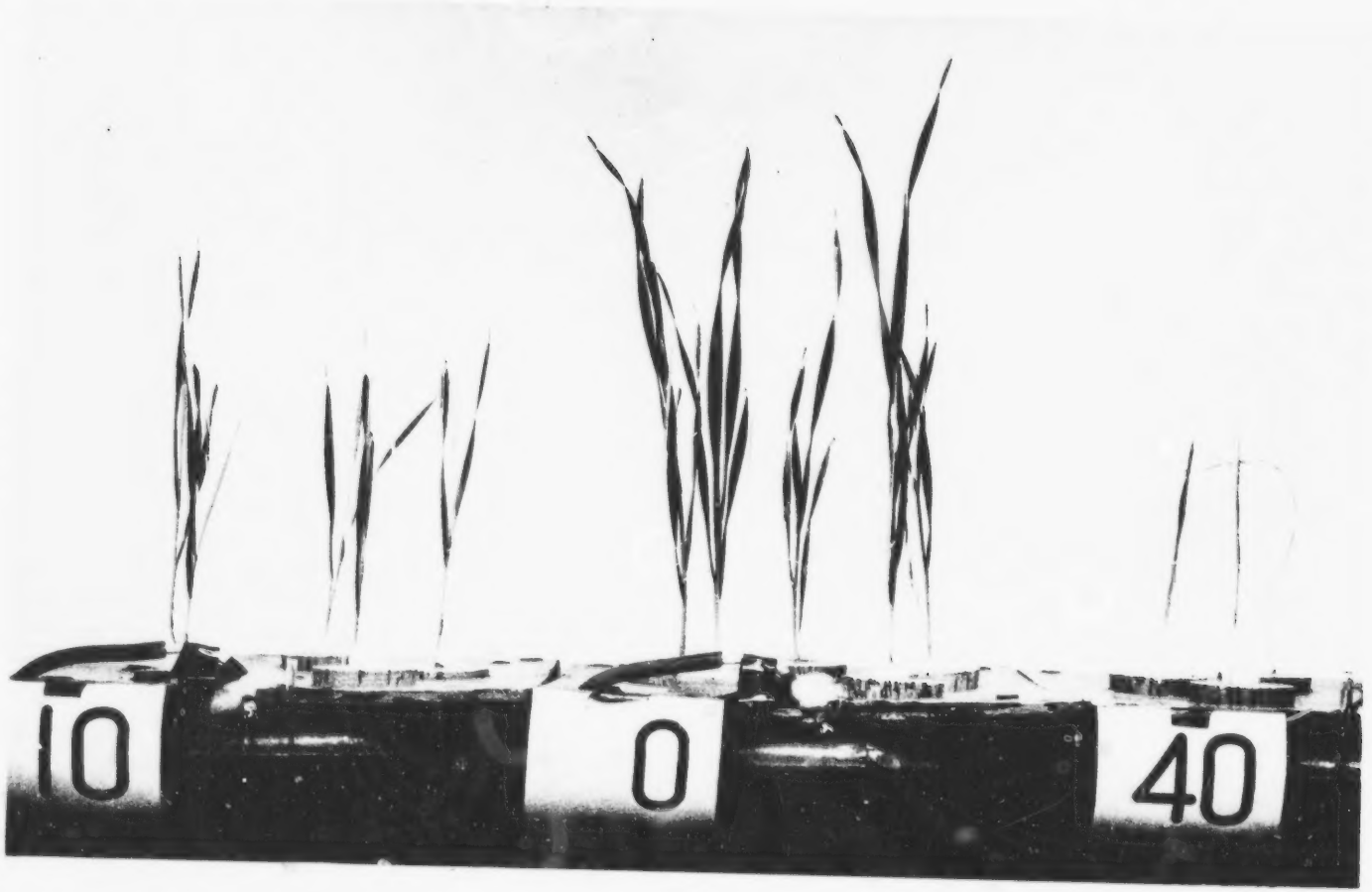


FIGURE 19. BARLEY GROWN IN 0, 10 AND 40 MG/L ABS WATER CULTURE.

Results of radioassay reported in Table 10 again showed a predominance of ABS in the roots and a higher percentage of ABS degradation products in the stems and leaves. Unlike the sunflower experiment, however, barley uptake of ABS increased almost proportionately to increased ABS concentration in the water culture.

TABLE 10

AVERAGE UPTAKE OF ABS AND S BY BARLEY GROWN IN WATER CULTURE

| Water Culture ABS, mg/l | Plant Part | S, µg/g dry weight | ABS, µg/g dry weight | Degradation of ABS, % |
|---|---|---|---|---|
| 10 | Stems and leaves | 310 | 140 | 55 |
| 10 | Roots | 4,800 | 4,100 | 14.6 |
| 40 | Stems and leaves | 1,500 | 940 | 37.3 |
| 40 | Roots | 20,000 | 20,000 | 0 |

Plant Growth in Soil

Sunflower plants germinated in vermiculite were grown in Yolo Sandy Loam contained in 7-inch flower pots. Drainage was provided by an underdrain of Fairoaks pea gravel overlaid with Monterey coarse and Monterey fine sand. Five solutions (tap water, settled sewage, and concentrations of 5, 10, and 50 mg/l ABS in tap water) were each applied to five replicate pots for a period of 57 days. These solutions all contained tracer $ABS^{35}$ and were administered in 1-liter quantities twice a week. No fertilizer was added to the soil, nor were any nutrients added to the solution.

Under these conditions, the plants fed sewage effluent flourished in comparison to those receiving tap water, as seen from their growth curves (Figure 20). This demonstrates that the beneficial effects of

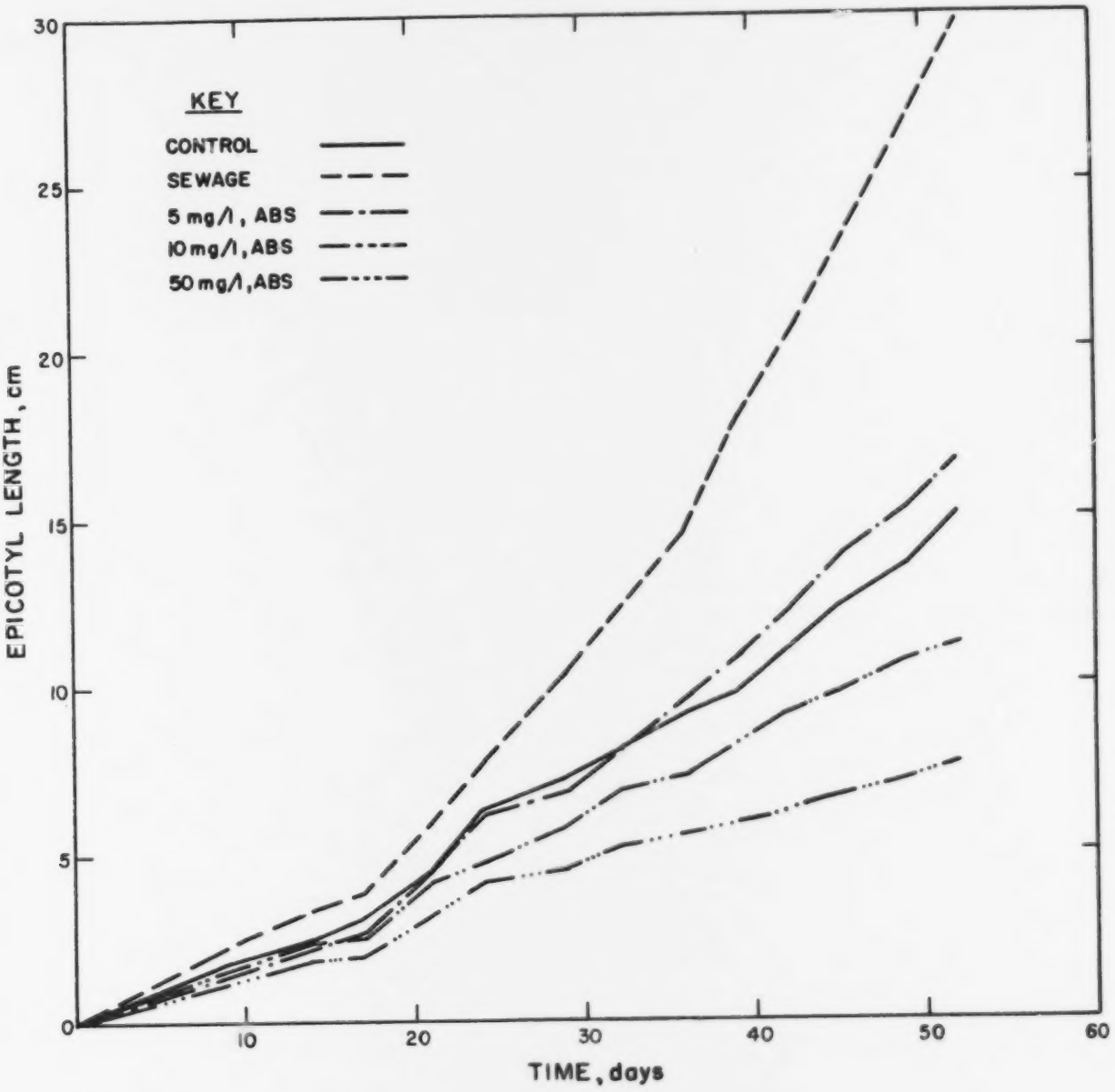


FIGURE 20. GROWTH OF SUNFLOWERS IN SOIL

nutrients outweigh the harmful effects of ABS in sewage for plants grown in a nutrient-deficient soil. As found in the water-culture studies, plants grown at 10- and 50-mg/l ABS were retarded. Figure 21 depicts pictorially a typical plant grown at each ABS concentration, from which it is evident that retardation occurring at 10- and 50-mg/l ABS is not as severe in soils as in water cultures. Although the 5-mg/l plants grew slightly taller than the controls, Table 11 shows that the control plants averaged slightly more fresh weight. Neither of these differences was statistically significant, however.

TABLE 11

EFFECT OF ABS ON SUNFLOWERS GROWN IN SOIL

| ABS in Water, mg/l | Mean Weight, g | | Mean Water Content, Fresh Weight Basis, % |
|---|---|---|---|
| | Fresh | Dry | |
| 0 | 3.72 | 0.80 | 78.5 |
| 5 | 3.62 | 0.75 | 79.2 |
| 10 | 2.61 | 0.58 | 77.5 |
| 50 | 1.23 | 0.33 | 73.0 |
| Sewage* | 26.96 | 5.29 | 80.5 |

*ABS concentration in sewage ranged from 4.6 to 12.7 mg/l.

Plant analyses reported in Table 12 show less ABS uptake (measured only in the stems and leaves) than was the case when plants were grown in water culture (51.5 per cent less at 10 mg/l and 96.1 per cent less at 50 mg/l). In both soil and water cultures, at 10-mg/l ABS the uptake by the plants of S related to ABS was approximately the same, but breakdown of ABS was almost double in the plants grown in soil. This suggests that the extended growing period allows more time for breakdown

FIGURE 21. SUNFLOWERS GROWN IN SOIL AT VARIOUS ABS CONCENTRATIONS.

of ABS to occur both in soil and in the plant. Another factor appears to be a difference in the mechanisms of ABS uptake by plants grown in the two media. At 50-mg/l ABS, plants grown in soil contained only 39.3 per cent as much ABS as the plants grown at 40-mg/l ABS in water culture, and had but 17.4 per cent as much ABS related S as the latter. From these results it is evident that ABS toxicity studies in water culture greatly exaggerate adverse effects on plant growth.

TABLE 12

UPTAKE OF ABS AND S BY SUNFLOWERS GROWN IN SOIL

| ABS in Water, mg/l | Mean ABS in Soil, µg/g | Mean S in Plant, µg/g dry weight | Mean ABS in Plant, µg/g dry weight | Mean Degraded ABS in Plant, % |
|---|---|---|---|---|
| 5 | 7.3 | 165.0 | 34.2 | 79.4 |
| 10 | 12.4 | 186.0 | 43.0 | 77.0 |
| 50 | 61.0 | 719.0 | 93.5 | 87.0 |
| Sewage* | 17.7 | 247.0 | 79.2 | 67.9 |

*ABS concentration in sewage ranged from 4.6 to 12.7 mg/l.

Despite the decrease in plant ABS-uptake noted in soil studies, this uptake is still sizable enough to be of significance in reducing the ABS content of soils. For example, the sewage-fed plants took up an average of 247.0 µg of ABS-related S per g dry weight of plant material (stems and leaves). The average dry weight of these plants was 5.29 g, giving a total uptake in the stems and leaves of 1,304 µg. This is 3.3 per cent of the ABS adsorbed on the soil. An estimate of root ABS uptake at double this figure (water culture studies showed a root-to-stem ABS concentration ratio of approximately 10-to-1 for 10-mg/l sunflowers, and a weight ratio of approximately 1-to-5) would mean that these plants might remove ten per cent of the ABS adsorbed on the soil.

The results of the sunflower experiment indicated a need for separation of the two critical experimental factors in plant growth: nutrients and ABS. This was accomplished in the present experiments with barley as the test plant by applying water solutions of ABS to one group of barley plants, and settled sewage plus ABS to another group. From the sunflower experiment it was also evident that the soil, Yolo sandy loam, was deficient in nutrients. Chemical fertilizers were therefore added, following the method of J. Vlamis (6). Addition of 0.457 g ammonium nitrate, 0.426 g primary calcium phosphate, and 0.296 g potassium sulfate was made to 1,600 g of soil in each pot. Nine barley seeds were planted per pot, and six days later the four smallest seedlings were removed. The first group of twenty-five pots was divided into replicate groups of five, and watered twice a week with distilled water containing 0, 5, 10, 15, and 20 mg/l ABS. A second group of twenty pots, also divided into replicate groups of five, received settled sewage containing added ABS in concentrations of 0, 5, 10, and 15 mg/l. Tracer $ABS^{35}$ was added to all pots. The plants were grown in direct sunlight for a period of 57 days.

There was no significant difference in plant height or plant weight between the control and test plants of each group. Comparing the two groups, however, the plants receiving settled sewage were 1.2 times taller, 2.8 times heavier, and yielded 2.3 times more grain than those receiving water. Therefore, barley was found to be insensitive to ABS, but responsive to nutrients in sewage over and above normal fertilization practices.

The barley grain was harvested and analyzed for ABS uptake. As shown in Table 13, the maximum ABS concentration was only 6.8 $\mu g/g$ and

three samples had no ABS. An average of 97.1 per cent of the radiosulfur found in the grain was related to intermediate breakdown products of ABS.

TABLE 13

AVERAGE CONCENTRATION OF ABS AND S IN BARLEY GRAIN
(Barley Grown in Soil)

| ABS Concentration, mg/l | S, µg/g dry weight | ABS, µg/g dry weight | Degradation of ABS, % | Degradation of ABS to Inorganic, % |
|---|---|---|---|---|
| In Water: | | | | |
| 5 | 59 | 0 | 100.0 | 1.9 |
| 10 | 41 | 0.6 | 98.5 | 2.0 |
| 15 | 115 | ---* | ---* | 2.8 |
| 20 | 92 | 6.8 | 92.8 | 4.3 |
| In Settled Sewage:† | | | | |
| 0 | 37 | 4 | 89.0 | 2.4 |
| 5 | 67 | 0 | 100.0 | 2.1 |
| 10 | 72 | 0.6 | 99.2 | 1.2 |
| 15 | 94 | 0 | 100.0 | 0 |

*Sample lost.

†Average concentration of ABS in settled sewage was 6.4 mg/l.

Lupinus Albus in Soil

A third study in soils was made with Lupinus albus. The procedure was similar to the barley experiment except that all plants were irrigated with distilled water and no tracer was added. ABS concentrations of 0, 5, 10, 15, and 20 mg/l were added to five replicate pots. The plants were grown in artificial light for thirty-five days after the initiation of ABS into the feed solution.

The results showed no significant difference between the control and the test plants in number of leaves or length of petioles.

Effect of ABS on Germinating Seeds

An investigation was made to determine whether ABS, like many toxic substances, has a stimulatory effect at concentrations below its toxic threshold. Studies were carried out using germinating seeds of Lupinus albus. The use of these seeds for studying the effects of pharmacological products has been recommended by F. Murayama (6) and the procedure which she developed was used in the present investigation.

Lupinus albus seeds were soaked in distilled water in the dark for 24 hours. The swollen seeds were then planted in vermiculite flats and incubated in a dark, water-saturated incubator at 20°C for 2-1/2 days. By this time the seeds had put out a root which had a hypocotyl length of about 15 to 35 mm. The hypocotyl was clearly defined and it was easy to measure its length with a millimeter scale.

Seeds with hypocotyl lengths between 21 and 30 mm were selected for the experiment. A yield of from 40 to 70 per cent of seeds of the desired hypocotyl length was obtained. All transfer operations and test incubations were carried out in artificial light to prevent photosynthesis.

Hoagland's solution (1/4 strength) (8) was made up and the desired amount of ABS added. The solution was aerated by vigorous shaking, then poured into small test tubes. There were 35 tubes at each ABS concentration. The seedlings were withdrawn from the vermiculite, washed in distilled water, measured, and placed in the small test tubes so that the complete hypocotyl was immersed in the test solution. The hypocotyl lengths of each group of seedlings was matched so that their average initial lengths fell within 0.1 mm.

The batches of seedlings were incubated for 24 hours at 20°C in a water-saturated atmosphere. After incubation the hypocotyl lengths were

measured and the percentage of growth calculated for each seedling. Three groups of 35 seedlings were grown in nutrient solution containing 0-, 2-, and 5-mg/l ABS.

At 0-mg/l ABS there was a 99.3 per cent increase in hypocotyl length after 24 hours; at 2-mg/l ABS, a 101.6 per cent increase; and at 5-mg/l ABS, a 112.1 per cent increase. The mean growth percentage at 2- and 5-mg/l ABS was compared statistically with the growth at 0-mg/l ABS (9). There was no significant difference between the 0- and 2-mg/l growth average, but the seedlings in 5-mg/l ABS showed a highly significant (99.75 per cent level) growth increase over the controls.

The results of this experiment supported the observed effect of 5-mg/l ABS in the unfertilized pot studies on sunflowers. However, when a second Lupinus albus run was made with concentrations of 0-, 5-, 10-, and 20-mg/l ABS in the test solutions, it was found that a significant growth stimulation was evident at all concentrations (Table 14). Moreover, the growth stimulation of ABS shows an increase with increasing ABS concentration.

TABLE 14

EFFECT OF ABS ON GERMINATION OF LUPINUS ALBUS SEEDS

| ABS Concentration, mg/l | Hypocotyl Growth, % |
|---|---|
| 0 | 80.2 |
| 5 | 93.4 |
| 10 | 90.3 |
| 20 | 99.1 |

It must be concluded, at this stage of growth of Lupinus albus, that ABS in concentrations from 5 to 20 mg/l has an increasingly stimulating effect.

## SUMMARY AND CONCLUSIONS

Two species of plants (sunflowers and barley) were grown in water culture at ABS concentrations of 0, 10, and 40 mg/l, and three species (sunflower, barley, and Lupinus albus) in soil at ABS concentrations up to 50 mg/l to determine the effect of ABS on plant growth and the uptake of ABS by plants. In soil studies, water solutions of ABS were compared to primary sewage effluents containing added amounts of ABS on both fertilized and unfertilized soils.

The major findings of these experiments were that although ABS caused severe growth inhibition in water culture studies (about 70 per cent at 10-mg/l ABS and almost 100 per cent at 40-mg/l), only one species (sunflower) of the three species grown in soil was adversely affected. Furthermore, plants irrigated with sewage containing from 4.6- to 12.7-mg/l ABS far surpassed in growth those irrigated with water, regardless of soil fertilization practices or the addition of up to 15-mg/l ABS to sewage. It may be concluded, therefore, that irrigation with sewage is beneficial to plants in spite of the presence of ABS in any amount likely to occur in sewage at the present time. Other notable findings of these studies are as follows:

1. In water culture studies, ABS concentrates to a considerable degree in plant roots (up to 9 per cent in sunflowers and 2 per cent in barley, on a dry weight basis) and to a lesser extent in stems and leaves (up to 0.2 per cent).

2. In water culture, degradation products of ABS were present primarily in the stems and leaves (up to 0.2 per cent) with only traces present in the roots.
3. Chlorosis of the leaves was evident in water culture studies--particularly in sunflowers grown in 10-mg/l ABS.
4. In soil studies, sunflowers grown at 5-mg/l ABS were unaffected, but those at 10-mg/l were 30 per cent retarded, and at 50-mg/l were 65 per cent retarded.
5. Sunflowers receiving primary sewage effluent (containing from 4.6- to 12.7-mg/l ABS) flourised in comparison to control plants receiving tap water and grown in unfertilized soil (7-fold increase in mean fresh weight).
6. Concentration of ABS in sunflowers grown in soil was small (less than 0.1 per cent of stem and leaf weight) and ABS degradation products were present in slightly greater concentrations (from 0.02 to 0.07 per cent of stem and leaf weight).
7. On the order of 10 per cent of the final soil ABS content was taken up by sunflowers (roots plus stems and leaves) irrigated with sewage effluent.
8. In soil studies, the only plants to be affected by ABS were sunflowers grown in unfertilized soil. The other two species (Lupinus albus and barley), grown in fertilized soil, showed no sensitivity to ABS up to 20 mg/l. It may then be that plant growth is retarded by ABS only in a nutrient-deficient soil.
9. Only a trace of ABS was found in the edible portion of barley irrigated with up to 25-mg/l ABS.
10. In the seed germination stage, Lupinus albus was increasingly stimulated by the presence of ABS in concentrations ranging from 5 to 20 mg/l.

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